



# **DNA DATABASE MANAGEMENT REVIEW AND RECOMMENDATIONS**

**ENFSI DNA Working Group  
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# 1 Introduction

This document discusses the different aspects of forensic DNA database management and makes recommendations, where deemed useful. Questions, remarks and additions in relation to this document can be sent to Izanda Puncule (izanda.puncule@ekspertize.vp.gov.lv). The first (2008) version of this document was approved at the 28<sup>th</sup> ENFSI DNA Working Group meeting which was held on 23rd - 24th April 2008 in Prague. Every year until 2017 an updated version of the document was presented at the ENFSI DNA Working Group meeting and republished on the ENFSI website after the approval of the group, from 2017 updates occur every two years.

The initial version of the document was produced with financial support from the ISEC Program of the European Commission - Directorate General Justice and Home Affairs as part of project JLS/2007/ISEC/506: "Improving the efficiency of European DNA data exchange".

This document was extensively reviewed for the 2019 version following feedback from 21 operational Databases using the audit trail contained in Appendix 2. Forensic Science Ireland as Co-Chair of the group coordinated this work and was supported by Dr. Siobhan Smith of FSI.

## 2 Establishing a forensic DNA database

The power of a forensic DNA database is that it can assist in the investigation of crimes by linking DNA profiles from crime-related biological trace material to each other and to the possible donors (or their relatives). Over the past 20 years, forensic DNA databases have proven to be very powerful in this respect. In spite of this success, not all ENFSI member countries have a DNA database yet.

The Council of the European Union invited its member states to consider establishing DNA databases<sup>1</sup> back in 1997. In 2001, a European Standard Set (ESS) of loci was established to enable the comparison of DNA profiles from different countries<sup>2</sup> and in 2009, the ESS was expanded with 5 extra loci<sup>3</sup>. In June 2008, the Council of the European Union converted the Treaty of Prüm into EU legislation (The EU Prüm Decision). The new EU legislation requires every EU Member State to establish a forensic DNA database and to make this database available for automated searches by other EU Member States. As DNA profiles are regarded as personal data, national privacy legislation, previously derived from the European Data Protection Directive 95/46 but, as of May 2018, derived from Regulation (EU) 2016/679 (General Data Protection Regulation) or, depending on the status of the institution conducting DNA analysis, Directive (EU) 2016/680 (Law Enforcement Directive), also applies to forensic DNA databases. This has certain consequences, which will be explained in chapter 14. It is therefore preferable to have specific DNA database legislation. The DNA Working Group of the ENFSI strongly feels that every European country should have a forensic DNA database to enhance:

- # the possibility of solving crimes
- # the number of crimes that are solved
- # the speed with which crimes are solved
- # the time that police can spend on other work
- # the possibility to link unsolved crimes
- # the possibility to identify false identities

The purpose of a national DNA database is usually defined in the legislation (e.g. intelligence tool, evidence provider, combat volume crime, combat serious crime, identify donors of stains, link crime scenes, etc.). This defined scope determines which categories of individuals should be included in the national DNA database.

### **ENFSI recommendation 1**

Every EU/ENFSI country should establish a forensic DNA database and pass specific legislation for its implementation and management.

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<sup>1</sup> EU Council Decision of 9 June 1997 on the exchange of DNA analysis results

<sup>2</sup> EU Council Resolution 9192/01

<sup>3</sup> EU Council Resolution 2009/C 296/01

### 3 Inclusion criteria

There are several criteria to consider in order to determine whether a DNA profile can/will be included in a DNA database. In the paragraphs below, these criteria are discussed.

#### 3.1 Source of the DNA profiles

In most countries with a DNA database, specific DNA legislation regulates which DNA profiles can or should be included in that DNA database. Some countries additionally require the specific authorization of a magistrate. Because the purpose of a DNA database is to find matches between crime-related stains and persons, these two types of DNA profiles are almost always present in a DNA database.

##### Crime-related stains

These are the DNA profiles which are assumed to originate from the perpetrators of crimes. It is the responsibility of the police to collect crime-related items. When the origin of the trace is unclear, reference samples (e.g. from the victim or from witnesses) should be collected, and their DNA profiles should be compared to those of the crime-related specimens to prevent DNA profiles from innocent people being included in the DNA database. DNA testing in high-volume crime (burglaries, etc.) is often very standardized and automated, to increase the number of traces analyzed and to decrease the throughput time from crime scene to inclusion in the DNA database. Specimens taken at these types of crime scenes should be chosen in such a way that the possibility that they originate from a perpetrator is maximized. Examples of such “safe” traces are: bloodstains (e.g. on broken windows), saliva stains (e.g. on tins, cups, bottles), cigarette butts and chewing gum, regarding which the residents of the burgled house can testify that they did not produce those samples.

Usually the types of crime from which stains originate correspond with the types of crime for which persons can be forced to provide a DNA sample. However, in some countries, there are no limitations with regards to the types of crime from which stains can be included in the DNA database. In practice, stains related to minor crimes are not collected due to the priority given to more serious crimes, but the absence of limitations on crime scene stains opens up the possibility of solving minor crimes (like littering or damaging public or private property), if the individual corresponding to the stain has already been included in the DNA database for a more serious crime. Moreover, linking minor to more serious crimes may yield additional investigative information which may speed up investigation of the more serious crime.

##### **ENFSI recommendation 2**

The type of crime-related stain DNA profiles which can be included in a DNA database should not be restricted.

##### Persons

Several categories of persons may be included in a DNA database.

- Convicted persons, persons who have been found guilty of a crime by a court of law and may (or may not) be (conditionally) convicted to imprisonment, a penalty, labor, hospitalization or a combination of these. A conviction can be overturned by a successful appeal to a higher court. In some countries it is possible to include persons in the national DNA database who have been convicted in the past and who have already completed their imprisonment. This is called retrospective sampling.
- Suspects, persons who have not yet been found guilty but are officially the subject of investigation and/or prosecution.
- Arrestees, persons who have been taken into custody by the police but are not (yet) a suspect as defined above.
- Volunteers, persons outside the above-mentioned categories who have agreed to give a DNA sample for investigative purposes. In some countries, volunteers can also be included in the national DNA database with their consent.

The legal criteria for the inclusion of convicts, suspects and arrestees in a national DNA database are usually either specific types of crime or the maximum punishment that the law allows for a crime.

Obtaining a DNA sample from convicted persons, suspects and arrestees may involve several steps.

- A person may first be asked to give a sample on a voluntary basis;
- An official police or judicial order may be served to provide a sample, either directly or upon refusal to give the sample on a voluntary basis;
- Various actions are possible in different countries upon refusal to provide a sample: conviction for the refusal, physical force to obtain a sample, or taking a sample from an object with the person's cell material (a surrogate sample). A conviction for the refusal does not result in the production of a DNA profile (and the inclusion of the DNA profile in the national DNA database) and hence is not a logical measure in DNA database legislation.

Since the identification of the donor of a stain depends on the presence of the donor in the DNA database, more donors can be identified if more relevant persons are included in the DNA database. Moreover, the persons included in the DNA database should adhere to the scope of the DNA database. For instance, including high volume crime scene stains but only persons convicted of sexual and capital crimes will not produce many matches.

### **ENFSI recommendation 3**

To increase the chance of identifying the donors of stains, the number of persons in a DNA database who are likely to be the donors of those stains should be as large as legally (and financially) possible.

Apart from nationally collected DNA profiles, DNA profiles originating from international legal comparison requests may also be included, to enable repeated comparisons against newly added DNA profiles. See also: chapter 22.

#### Victims

Some countries allow the inclusion of DNA profiles from the deceased victims of unsolved crimes in their DNA database. The purpose of this is to find matches which may help to solve the crime. If, for instance, the DNA profile of a dead victim who was stabbed to death later matches a blood stain on a knife, then the owner of the knife may become a murder or a manslaughter suspect.

There are two types of victims: identified and unidentified victims. Unidentified persons who are not apparent victims of a crime are usually included in a missing persons DNA database, but may be compared with the criminal DNA database in an attempt to identify them (see chapter 23). The "risk" of including victims is getting matches with other unsolved crimes, in which case the victim becomes a suspect. Therefore, victims who are still alive, like other volunteers, should be informed and asked to give their consent.

#### Missing persons

Some countries allow the inclusion of DNA profiles from missing persons if there is a suspicion that a crime is involved in their disappearance. The purpose of this is the same as for the inclusion of victims, namely, to find matches which may help to solve the crime.

#### Elimination profiles

The inclusion of DNA profiles in the DNA database for contamination detection purposes is dealt with in chapter 4.5.

## **3.2 Choice of loci**

Most countries use commercially available kits to produce DNA profiles for inclusion in their DNA databases. Table 1 shows the contents of the different kits which are or have been commercially available, as well as the composition of the different standard sets discussed below. Some kits are included which are no longer sold commercially (e.g. QUAD, SGM). Historically, these kits were used in the creation of the first DNA databases, but their discriminating power is insufficient to generate meaningful matches in relation to the millions of DNA profiles available for comparison today.

The EU Council resolutions 2001/C 187/01 and 2009/C 296/01 call upon European countries to use the European Standard Set (ESS) as a minimum to enable the international comparison of DNA profiles. In the USA, the required number of loci for the inclusion of a reference profile in the national DNA database of the USA (CODIS) used to be 13, but in 2015, the CODIS core locus set was expanded to 20 loci<sup>4</sup>. The INTERPOL Standard Set of Loci (ISSOL) is equal to the European Standard Set, plus the Amelogenin locus. Until December 2009, the European Standard Set of Loci contained only 7 loci. This was enough for occasional exchanges of DNA profiles between countries.

However, when massive exchanges of DNA profiles are undertaken, as has been made possible by the INTERPOL DNA database and the EU Prüm Decisions, 7 loci are generally insufficient, because the chance of adventitious matches becomes significant, and makes a routine process inefficient. In addition, each DNA database contains a significant portion of partial profiles with a much higher probability of matching randomly. This is why ENFSI has recommended that the European Standard Set of Loci should be expanded by 5 additional loci and the Council of the European Union adopted this recommendation on 30 November 2009. In the meantime, commercial companies have produced kits which contain these new loci to enable the implementation of the new ESS loci.

The locus D5S2500 is contained in the Investigator HDplex Kit of Qiagen, the 21+1 kit of AGCU Scien Tech and the Goldeneye DNA ID 22NC kit of Peoplespot. It has been shown however that the D5S2500 locus in the 21+1 kit of AGCU Scien Tech is incorrectly typed and actually is located 1643 nucleotides away from the correct D5S2500 locus and may be called D5S2800<sup>5</sup>. The D5S2500 locus in the Investigator HDplex Kit of Qiagen is correctly typed. For the D5S2500 locus in the Goldeneye DNA ID 22NC kit of Peoplespot no data were found to verify its typing.

The frequencies of the alleles of the different loci for different populations can be found in different sources, which are summarized in a publication of the DNA Commission of the ISFG<sup>6</sup>.

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<sup>4</sup> D.R. Hares (2015) *Forensic Science International: Genetics* 17 (2015) 33–34. Selection and implementation of expanded CODIS core loci in the United States

<sup>5</sup> C. Phillips et al (2016) *Forensic Science International: Genetics* 23 (2016) 19–24. D5S2500 is an ambiguously characterized STR: Identification and description of forensic microsatellites in the genomics age

<sup>6</sup> M. Dodner et al (2016) *Forensic Science International: Genetics* 24 (2015) 97–102. Recommendations of the DNA Commission of the International Society for Forensic Genetics (ISFG) on quality control of autosomal Short Tandem Repeat allele frequency databasing (STRidER)



Company	Thermo Fischer	Promega	Qiagen	Gordiz	Peoplespot	AGCU Scien Tech	Historic kits	Standard sets
Chromosome								
Locus								
1	F13B							
	D1S1627							
	D1S1656							
	D1S1677							
	D1GATA113							
2	D2S1338							
	TPOX							
	D2S441							
	D2S1360							
	D2S1776							
3	D3S1358							
	D3S1744							
	D3S3045							
	D3S4529							
4	FIBRA (FGA)							
	GABA							
	D4S2366							
	D4S2408							
5	D5S818							
	CSF1PO							
	D5S2500							
6	F13A							
	ACTBP2 (SE33)							
	D6S1043							
	D6S474							
	D6S477							
	D6S1017							
7	D7S820							
	D7S1517							
	D7S3048							
8	D8S1179							
	LPL							
	D8S1132							
9	Penta C							
	D9S1122							
10	D10S1248							
	D10S2325							
	D10S1435							
11	TH01							
	D11S2368							
	D11S4463							
12	WA							
	CD4							
	D12S391							
	D12ATA63							
13	D13S317							
	D13S325							
14	D14S608							
	D14S1434							
15	FES/FPS							
	Penta E							
	D15S659							
16	D16S339							
17	D17S1290							
	D17S1301							
18	D18S51							
	D18S535							
	D18S853							
19	D19S433							
	D19S253							
20	D20S482							
21	D21S11							
	Penta D							
	D21S2055							
22	D22S1045							
	D22GATA198B05							
XY	Amelogenin							
Y	DYS19							
	DYS385a/b							
	DYS389I							
	DYS389II							
	DYS390							
	DYS391							
	DYS392							
	DYS393							
	DYS437							
	DYS438							
	DYS439							
	DYS448							
	DYS456							
	DYS458							
	DYS481							
	DYS529a/b							
	DYS570							
	DYS576							
	DYS635							
	DYS643							
	GATA-H4							
	Y Indel							
Total nr of loci	11	10	10	7	12	3	16	8
	16	16	16	17	15	24	9	9
	16	16	16	17	17	17	17	14
	18	18	18	7	5	21	24	27
	5	21	24	27	16	17	12	14
	7	14	14	7	3	3	13	9
	3	13	23	9	11	20	20	22
	18	18	18	18	22	22	22	20
	22	22	22	22	22	22	22	22
	26	26	26	31	9	9	9	9
	9	9	9	9	9	9	9	9
	33	33	33	33	33	33	33	33
	4	4	4	4	4	4	4	4
	4	4	4	4	4	4	4	4
	12	12	12	12	12	12	12	12
	24	24	24	24	24	24	24	24
	21	21	21	21	21	21	21	21

### 3.3 Number of loci

For the comparison of DNA profiles between EU countries, DNA profiles must comply with the Prüm inclusion rules. For comparison of DNA profiles within a single country, however, other criteria may apply. DNA profiles from crime scene stains may not contain all loci present in the kit(s) used in a country to produce DNA profiles. These partial DNA profiles may be included in national DNA databases, provided they have a high enough evidential value and/or the chance of producing adventitious matches is not too high (see chapter 7). Two criteria commonly used for the inclusion of partial profiles are 1) minimum number of loci and 2) maximum random match probability. The second criterion is better because a DNA profile containing only 4 or 5 loci may have a lower random match probability than a DNA profile containing 6 loci if (some of) the alleles in the former are rare.

A simulation study has been published, which shows the influence of including DNA profiles with lower numbers of loci on the number of genuine and adventitious matches, generated in a simulated Swiss DNA database<sup>7</sup>.

#### **ENFSI recommendation 4**

Managers of national DNA databases should establish (together with other stake-holders) criteria for the inclusion of partial DNA profiles to obtain an acceptable balance between the minimum allowable level of evidential value (maximum random match probability) of a DNA profile and the maximum number of adventitious matches a partial DNA profile is expected to generate.

Sometimes an unsolved crime is so serious that a DNA profile which does not meet the minimum criteria for inclusion in the national DNA database is still searched against a national DNA database, accepting the fact that many of the matches found will be adventitious matches. Tactical police work is then necessary to find out if any of the matches lead to a potential suspect. If no potential suspect is found by the police, the search action may be repeated after some time or at regular intervals, because new people will have been added to the national DNA database. The CODIS autosearcher mode produces only the new matches in these types of search actions, which saves work in sorting out old and new matches.

For historic reasons, the countries who started early with their DNA databases (like the United Kingdom and the Netherlands) still have DNA profiles in their DNA databases which were produced by the older commercial kits like QUAD (4 loci) and SGM (6 loci + Amelogenin). For economic reasons, these DNA profiles are often only upgraded when they produce a match. This also implies, however, that these profiles often do not fulfill the criteria for international comparison, which is a missed chance to solve the case from which the DNA profile originates. An upgrade of a DNA profile is, of course, only possible if the cell material or the DNA extract is still available for further testing.

**ENFSI recommendation 5** If possible, DNA profiles should be upgraded after a match in the national DNA database if it increases the evidential value of the match and decreases the possibility of an adventitious match.

The number of loci in reference samples should be the maximum number of loci present in the kit(s) used for the production of the DNA profiles of reference samples, to increase the chance of finding relevant matches with partial DNA profiles. However, sometimes this is not possible due to an allelic drop out or a true or apparent trisomy.

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<sup>7</sup> T. Hicks et al (2010) FSI Genetics 4(4) 232-238. Use of DNA profiles for investigation using a simulated national DNA database: Part I. Partial SGM Plus® profiles

**ENFSI recommendation 6** Reference sample profiles should preferentially be loaded to a database only if a complete profile (maximum number of loci) is obtained using the PCR chemistry of choice.

It goes without saying that the reliability of the matches produced in a DNA database is dependent on the reliability of the DNA profiles used in the match. A wrongly called allele may prevent a match and a sample mix-up may produce a false match. For these and other reasons, labs producing DNA profiles for DNA databases should be able to show objectively that they produce DNA profiles with quality-driven processes. This means, for example, that there must be arrangements in place whereby the laboratory can demonstrate:

- The validation of its analytical processes;
- Arrangements for continuous monitoring of data quality and consistency;
- Arrangements for error identification, error handling and incorporation of corrective and preventative actions

Council Framework Decision 2009/905/JHA of 30 November 2009 “on the accreditation of forensic service providers carrying out laboratory activities” requires ISO 17025 accreditation for all forensic DNA laboratories.

**ENFSI recommendation 7**

Labs producing DNA profiles for a DNA database should, as a minimum, be ISO-17025 (and/or national equivalent) accredited and should participate in challenging proficiency tests.

In some countries, laboratories which supply DNA profiles to the national DNA database are audited by the custodian of the DNA database. Apart from this, the custodian of the DNA database should have regular contact with the suppliers of the DNA profiles to exchange information about legal and technical developments, changes in the inclusion and matching rules, incidents, etc.

**ENFSI recommendation 8**

The custodian of the DNA database should have regular contact with the suppliers of the DNA profiles to exchange information about legal and technical developments, changes in the inclusion and matching rules, incidents, etc.

### **3.4 DNA profiles produced from low levels of DNA**

DNA profiles produced from low levels of DNA, whether by a standard or enhanced number of PCR-cycles or by signal enhancing techniques like increased injection settings or post-PCR clean-up, can contain allele drop-ins and allele drop-outs, even if a consensus profile is produced from repeated determinations<sup>8</sup>. Hence, they may never turn up matches when included in a DNA database, if all alleles are required to match. If DNA profiles produced from low levels of DNA are included in a DNA database, they should be recognizable and/or a dedicated match strategy (allowing one or more mismatches) should be used to detect possible allelic drop-ins and drop-outs (as will be discussed in § 5.4). For a discussion on mixed profiles from low levels of DNA, see § 3.9

**ENFSI recommendation 9** If a laboratory uses enhanced techniques to produce DNA profiles, they should be searched using a dedicated (near) match strategy.

### **3.5 Composite DNA profiles**

The smaller PCR products of DNA profiles from stains regularly show higher peak heights than larger PCR products. This is due to partial breakdown of the DNA. It can even occur that the larger PCR products

<sup>8</sup> C.C.G. Benschop et al. (2011) Low template STR typing: Effect of replicate number and consensus method on genotyping reliability and DNA database search results. *Forensic Sci Int Genet.* 5, 316-328.

disappear below the detection threshold, while the smaller PCR products still show good peaks. Sometimes the peak heights of the larger PCR products can be improved by increasing the input of the PCR reaction, but this can often result in the overloaded peaks of smaller PCR fragments. By using low as well as high input during the PCR reaction, two DNA profiles may be obtained, one with clear, legible peaks of the smaller fragments and the other with clear, legible peaks of the larger fragments. These can then be combined into a composite DNA profile. This should, however, only be done with DNA profiles obtained from the same DNA extract and not with DNA profiles obtained from different DNA extracts (even if they come from the same sample), because it cannot be excluded that different samples (or different parts of a sample) contain DNA from different individuals.

#### **ENFSI recommendation 10**

Composite DNA profiles should only be created from DNA profiles generated from the same DNA extract because it cannot be excluded that different extracts, even from the same sample, contain DNA from different individuals.

### **3.6 Rare alleles/chromosomal anomalies**

For each commercial kit, the known alleles of each locus and their relative frequency (in several different populations) is described in the manual of the kit. From time to time, new alleles are observed in DNA profiles and it is important to consider whether these new alleles should be included in the DNA database, and which frequency they should be assigned in order to calculate the probability of the particular DNA profile in the population of interest (i.e. the so-called random match probability). When a new allele is observed, its appearance should first be confirmed by repeated DNA extraction, PCR, capillary electrophoresis and allele calling. Before including the new allele in the DNA database, a literature search may be conducted to see whether the new allele has been observed and/or sequenced before. A good source for this is the DNA database of NIST<sup>9</sup>. If a new allele has not been sequenced yet, it can be sent to NIST for sequencing. Only new alleles whose size can be accurately determined using the internal DNA size standard should be included in the DNA database. An additional criterion for including a new allele in the DNA database is the number of internal or/and external observations of the new allele.

The relative frequency attributed to a new allele may be one divided by the size of the reference database used to estimate allelic proportions, a predetermined (low) relative frequency or a proportion calculated according to alternative statistical estimation procedures. Allelic relative frequencies can be estimated using methods like the Balding<sup>10</sup> size correction formula (i.e. a Bayesian estimator).

STRidER (**STRs for Identity ENFSI Reference Database**) is the expanded and enhanced version of the ENFSI STRbase (2004-2016). This curated online high quality STR allele frequency population database enables scientifically reliable **STR genotype probability estimates** and provides **quality control** of autosomal STR data. A suite of software tools has been developed at the Institute of Legal Medicine, Medical University of Innsbruck to scrutinize STR population data and thus increase the quality of datasets to ensure reliable allele frequency estimates. STRidER acts as **frequency database and software platform** for the development of novel tools for STR data QC and other forensic analyses, <https://strider.online/>.

#### **ENFSI recommendation 11**

When a new allele is observed in a DNA profile, its presence should be confirmed by repeated DNA extraction, PCR, capillary electrophoresis and allele calling of the entire DNA profile. Only new alleles whose size can be accurately determined using the internal DNA size-standard should be included in the DNA database.

Sometimes chromosomal anomalies are observed in DNA profiles. As a result, a locus may show more than 2 peaks. A well-known example of this is trisomy 21, which causes Down's syndrome. As these

<sup>9</sup> <http://www.cstl.nist.gov/biotech/strbase/index.htm>

<sup>10</sup> Balding, DJ (1995) Estimating products in forensic identification. J. Am. Stat. Assoc. 90:839-844.

chromosomal anomalies are rare and hence contribute to the evidential value of the DNA-profile, it would be logical to recommend that they be included in the DNA database. However, extra peaks can also be caused by somatic mutations, which may appear only in certain tissues/body fluids. This means that DNA profiles from different sample types (e.g. buccal swab and blood) may appear to be from different donors and might be dismissed as a match. This can, of course, contribute to the evidential value after the match has been found in the DNA database. However, if the default search strategy is moderate, a profile containing a trisomy will match a profile without the trisomy<sup>11</sup>.

**ENFSI recommendation 12** Alleles from loci with chromosomal anomalies may be included in a DNA database if the default search strategy is “moderate”. If the default search strategy is “high”, wild cards may be used, as long as an agreed set of wildcards is determined to permit meaningful international exchange.

Sometimes an apparent trisomy can occur when an unusually long or short allele of a locus falls out of its own bin and falls into a neighboring bin in the electropherogram. The allele calling software then calls three peaks in one bin and only one in the neighboring bin. This situation can be clarified by using a monoplex PCR or by using a different kit where the two loci involved are not adjacent on the electropherogram.

A regularly observed tri-allelic pattern for TPOX was recently analyzed<sup>12</sup>. The results showed that some of these tri-allelic patterns are caused by a translocation of allele 10 of the TPOX locus to the X-chromosome.

The inclusion rules for DNA profiles which are compared on the basis of the EU Prüm Council Decisions state that a tri-allelic locus should be converted into the first allelic value, plus a wildcard. This is in contrast with recommendation 12, but cannot be changed at this moment because Council Decision 2008/616/JHA, which contains the inclusion rules, will not be amended until all EU countries are operational.

### 3.7 Wildcards

If there is uncertainty about the presence or absence of an allele in a DNA profile, a so-called “wildcard” may be included in the DNA profile. This may be the case with low peaks, where the DNA analyst cannot determine whether it is a homozygote peak or a locus where one allele has dropped out.

In some countries, a wildcard is used to replace a rare allele which is not in the ladder-range of the DNA kit used. In this case, the wildcard represents a designated allele which can be used to verify a match with a DNA profile containing the same wildcard. Searching with wildcards means that any allele is accepted as a match for the wildcard allele. Different countries use different designations for their wildcards. For the purposes of international comparison, these national designations have to be converted into mutual designations. Countries that exchange DNA profiles under the terms of the EU Prüm decision presently use “\*” as a wildcard. There has been a proposal to use “\*” for a wildcard that represents a designated allele, and to use “B” for a wildcard that represents an unknown allele, but this proposal has not yet been implemented. The use of wildcards increases the chance of finding adventitious matches in the DNA database<sup>13</sup>, but if the wildcard represents a rare allele and both profiles prove to contain this rare allele, the evidential value of the match greatly increases.

### 3.8 Mixed profiles

Mixed profiles can occur when two or more individuals have left cell material on the same object (e.g. smoking from the same cigarette or drinking from the same bottle), or when, for example, cells of a perpetrator are mixed with cells of a victim (which often occurs in rape cases). If possible, mixed DNA

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<sup>11</sup> An inventory of tri-allelic pattern observations for the commonly used STR markers can be found at:

[http://www.cstl.nist.gov/biotech/strbase/tri\\_tab.htm](http://www.cstl.nist.gov/biotech/strbase/tri_tab.htm)

<sup>12</sup> Picanço, J.B. et al (2015) Identification of the third/extra allele for forensic application in cases with TPOX tri-allelic pattern. *Forensic Science International Genetics* 16, 88–93

<sup>13</sup> Tvedebrink, T. et al (2015) The effect of wild card designations and rare alleles in forensic DNA database searches. *Forensic Sci Int Genet* 16, 98-104

profiles should be interpreted and separated into their contributing DNA profiles. Mixed profiles from (known) victims and (unknown) donors occasionally can be resolved, because the alleles of the victim's DNA profile can be subtracted from the mixed profile. The remaining alleles must belong to the unknown donor. Mixed DNA profiles from two donors, however, can often only be completely designated into separate contributors if there is a significant difference in contribution between the two donors (a Major/Minor situation). A working group of the ISFG has produced a document with guidelines for the analysis of mixed profiles<sup>14</sup>. Several software tools, both commercial and open-source, have become available that can resolve mixtures and produce possible combinations of donor profiles (see the website of the ISFG <http://www.isfg.org/software> for open-source software). Such tools may be used, provided they are properly validated.

**ENFSI recommendation 13**

The guidelines in the document of the ISFG working group on the analysis of mixed profiles should be used for the analysis of mixed profiles. Software tools may also be used, provided they are properly validated.

In some DNA databases (like CODIS), mixed DNA profiles can be included and searched. This is very useful when a mixed DNA profile cannot be reliably resolved into its contributing components. In CODIS, it is even possible to designate the remaining alleles as “required”, if one of the participants of a mixed DNA profile has been identified. Matches with reference samples will only be shown if these required alleles are present in the reference sample DNA profile. A numerical match between a reference sample and a mixed profile must always be checked against the electropherograms of the DNA profile, because a numerical match may not be a true match, as shown in figure 1. For this reason, mixed profiles cannot currently be used for the automated international comparison of DNA profiles, like the comparisons which are performed under the terms of the EU Prüm Council Decision, and those conducted in the INTERPOL DNA database.

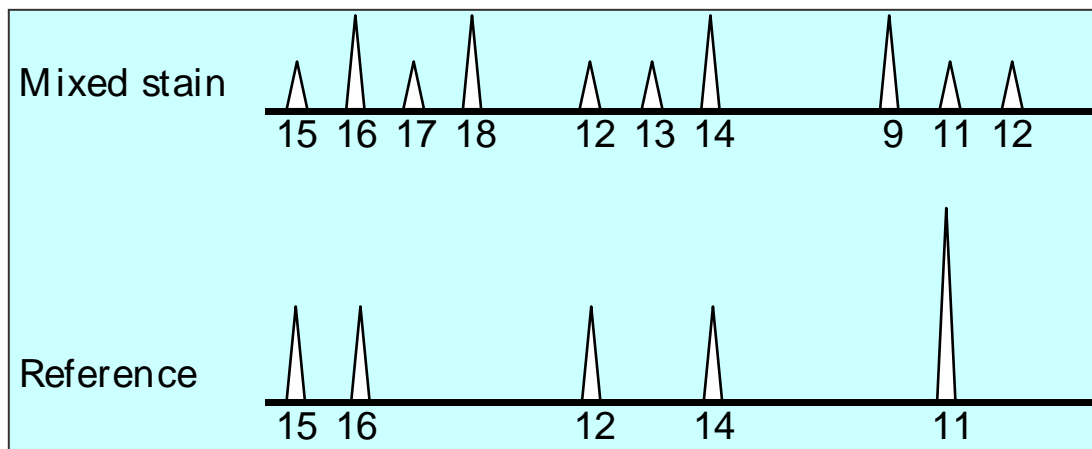


Figure 1: Three loci of a mixed stain and a reference sample which match on a numerical basis but are an unlikely combination when peak heights are taken into account

**ENFSI recommendation 14**

A numerical match between a reference sample and a mixed profile must always be checked against the electropherogram of the mixed profile.

Mixed profiles of more than 2 individuals should not be systematically included in a DNA database because they will generally produce too many adventitious matches. Manual searches using this type of profile may, however, be useful.

<sup>14</sup> Gill, P. et al (2006), 'DNA commission of the International Society of Forensic Genetics: Recommendations on the interpretation of mixtures', *Forensic Sci Int.* 160, 90-101

### **ENFSI recommendation 15**

Mixed profiles of more than 2 individuals should not be systematically included in a DNA database because they will generally produce too many adventitious matches.

Special software exists to resolve mixed DNA profiles into possible contributors (see above). These possible contributors can then be searched against the national DNA database of a country. Some people have expressed their concern that this will lead to an increase in false positive matches. Compared to the situation where mixed profiles themselves are included in a DNA database (which can, for instance, be done by countries using CODIS), conducting a search using the possible contributors of a mixed DNA profile will not lead to more false positive matches, provided that any resulting matches are interpreted with caution.

In rare cases, a mixed profile can be obtained from a single individual. This can happen when a buccal swab is taken from an individual who has received a bone-marrow or blood stem cell transplant in the context of medical therapy. As a result, the blood has the DNA profile of the tissue donor, whereas other body tissues still have the original profile of the individual. However, when taking a buccal swab, very small superficial blood vessels may be damaged, causing a mixed profile.

Mixed profiles obtained from low levels of DNA can contain allelic drop-in and drop-out peaks and are even more difficult to analyse than single-source profiles obtained from low levels of DNA. The use of consensus and composite profiles may assist in the analysis and interpretation of these profiles<sup>15</sup>. Special software has been developed to compare these profiles to reference samples, resulting in a likelihood ratio expressing the ratio of the probability of the results, given that the trace came from the person who is the source of the reference profile and one or more unknown persons; and the probability of the results, given that the trace originates from two (or more) unknown persons. LRmix Studio is an open-source example of such a software program<sup>16</sup>. SmartRank is a program that links LRmix Studio to a DNA database for the comparison of complex mixed profiles to all reference profiles in the DNA database<sup>17</sup>. This will result in a list of likelihood ratios for each reference profile in the DNA database. The names of the persons associated with the reference profiles with the highest likelihood ratios can then be used by the police as an investigative tool. Additional DNA testing may be necessary to confirm/reject that a candidate obtained in this way could be a true contributor to the mixed profile. SmartRank, developed with the support of an ENFSI Monopoly Grant, has been validated in 2017<sup>18</sup>. Many other programs, both commercial and open source (such as True Allele, STRMix, EuroForMix – this is not an exhaustive list) are now available. SmartRank<sup>15</sup>, STRMix<sup>19</sup> and Euroformix<sup>20</sup> can now compare mixed profiles to DNA databases<sup>21</sup>.

### **3.9 Sequence variation between STR alleles of similar size**

The present designation of STR alleles is based on the number of repeats, as determined by their size in capillary electrophoresis. More sensitive analyses using ion-pair reversed-phase high-performance liquid chromatography electrospray-ionization quadrupole time-of-flight mass spectrometry (ICEMS)<sup>22</sup>, or massive parallel sequencing<sup>23</sup>, have shown, however, that STR alleles in general display considerable sequence

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<sup>15</sup> C. Benschop et al (2013) Consensus and pool profiles to assist in the analysis and interpretation of complex low template DNA mixtures. *Int. J. Legal Med.* 127, 11-23.

<sup>16</sup> <http://lrmixstudio.org>

<sup>17</sup> <http://lrmixstudio.org/smartrank/>

<sup>18</sup> C. Benschop et al (2017) Validation of SmartRank: A likelihood ratio software for searching national DNA databases with complex DNA profiles. *Forensic Sci Int Genet.* 2017 Jul;29:145-153.

<sup>19</sup> <https://www.strmix.com/news/dblr-discussion-with-dr-maarten-kruijver/>

<sup>20</sup> <http://www.euroformix.com/?q=dnamatch2>

<sup>21</sup> The mentioning of trade names does not mean that ENFSI recommends or endorses any of these programs. The aim of ENFSI is to provide insight into what is available on the market.

<sup>22</sup> Oberacher et al. *Electrophoresis* 29 (2008) 23: 4739-50. The next generation of DNA profiling - STR typing by multiplexed PCR - ion-pair RP LC-ESI time-of-flight MS

<sup>23</sup> Børsting C, Morling N. Next generation sequencing and its applications in forensic genetics. *Forensic Sci Int Genet.* (2015) Feb 14. DOI: <http://dx.doi.org/10.1016/j.fsigen.2015.02.002>

variability, resulting in additional discrimination for alleles of identical size. In addition, the flanking sequences between the STR and the primer binding site show sequence variability. These findings will have significant consequences for forensic DNA typing:

- Alleles determined as similar by capillary electrophoresis will be differentiated due to sequence variability.
- Match probabilities will be lower than presently calculated, because allelic proportions will be smaller, resulting in the enhanced discrimination power of DNA typing, which is especially important for mixtures and partial DNA profiles.
- The established DNA databases can still be used, but the nomenclature of the alleles will have to be adjusted to deal with different alleles of similar size. The DNA commission of the International Society for Forensic Genetics (ISFG) has published considerations on minimal nomenclature requirements for massively parallel sequencing of forensic STRs<sup>24</sup>.

### **3.10 Non-autosomal STR markers**

In the previous paragraphs, only autosomal STR markers have been discussed. However, the X- and Y-chromosomes also contain STR markers. Y-chromosomal markers are especially important and are frequently used in forensic DNA testing because they can be used to reveal the presence of male DNA amongst an excess of female DNA. They can also help establish male familial relationships because they segregate unchanged as a haplotype from a father to his sons (provided there is no mutation). X- and Y-chromosomal markers can easily be stored in DNA databases like CODIS. The difference with other STR markers is that most Y-chromosomal STR markers contain only one allele due to their haploid nature. Searching with Y-chromosomal STR markers is also possible, but this implies a familial search which may need special permission from the competent authorities. With rapidly mutating Y-STRs, males of the same male lineage may still be distinguished from each other<sup>25</sup>.

#### **3.10.1 Y-chromosomal STR markers**

Y-chromosomal markers belong to the lineage (or haploid) markers. Due to the lack of recombination and the linear mode of inheritance, both the sampling strategy and the reporting of frequencies differs from autosomal DNA markers, but follows the same principles based on theories in population genetics and the laws of probability. Because of full linkage between markers within a Y-STR profile (haplotype), the product rule cannot be applied, and instead large haplotype reference databases are mandatory to perform calculations. The YHRD (Y-Chromosome Haplotype Reference Database) is the largest, annotated, strongly curated and quality-controlled forensic database<sup>26</sup>. It is designed to store haplotypes from hundreds of population samples from around the globe and to rapidly disseminate haplotype frequency data via the internet to forensic analysts. The databases also include several tools to analyze population substructure effects, to interpret matches between Y-STR profiles, to attach likelihood ratios in mixture analyses, and to formulate valid forensic testimonies. YHRD is built by direct submissions of population data from individual certified laboratories. Upon receipt of a submission, the YHRD staff examines the originality of the data, assigns an accession number to the population sample and performs quality assurance checks. The submissions are then released to the public database, where the entries are retrievable by search for haplotypes, populations, contributors or accession numbers. Currently the YHRD presents about 160,000 minimal haplotypes in 1015 different populations (Release 51, Feb. 2016). All population data published in forensic journals as FSI Genetics or International Journal of Legal Medicine are required to be validated by the YHRD custodians and are subsequently included in the YHRD<sup>27</sup>.

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<sup>24</sup> W. Parson et al. (2016) Massively parallel sequencing of forensic STRs: Considerations of the DNA commission of the International Society for Forensic Genetics (ISFG) on minimal nomenclature requirements. *Forensic Science International: Genetics*, Volume 22, May 2016, Pages 54-63

<sup>25</sup> Alghafri, R. et al. (2015) Single multiplex assay for simultaneously analyzing 13 rapidly mutating Y-STRs. *Forensic Sci Int Genet.* (2015) 17, 91-98

<sup>26</sup> Willuweit S, Roewer L (2007) Y-chromosome haplotype reference database (YHRD): update. *Forensic Sci Int Genet.* 1(2): 83-7

<sup>27</sup> Carracedo A. et al. (2014) Update of the guidelines for the publication of genetic population data. *Forensic Sci Int Genet.* 10, A1-A2



### 3.10.2 X-chromosomal STR markers

X-chromosomal STR markers can be useful in analyzing specific kinship cases. Like Y-chromosomal markers, they can be stored and searched in DNA databases like CODIS but familial search restrictions may also apply here. Furthermore, the particular linkage situation of the STR markers on the X-chromosome has to be taken into consideration in the case of biostatistical calculations<sup>28</sup>.

### 3.11 Amelogenin

Most commercial kits contain the amelogenin marker, which is present on both the X- and Y-chromosome. The amelogenin gene on the X-chromosome contains a 6 base-pair deletion, which results in different PCR fragment lengths and thus the ability to distinguish male and female DNA profiles. In rare cases, a mutation or a deletion in the amelogenin gene can result in the inability to produce a PCR-fragment which then gives a wrong impression about the sex of the DNA profile donor<sup>29</sup>. Because the amelogenin marker does not give foolproof results, some companies have added additional Y-chromosomal markers to their newest kits (e.g. Globalfiler, Powerplex Fusion).

### 3.12 Mitochondrial DNA (mtDNA) information

Mitochondrial DNA (mtDNA) information is frequently used in forensic DNA testing. The information is sequence-based and typically covers the mtDNA control region approx. 1 kb in length on the D-loop from positions 16024 to 576 (comprised of the hypervariable regions HV I-III). In contrast to autosomal DNA, of which only two copies are present in each cell, mtDNA is present in many hundreds of copies. For this reason, traces that fail to give an autosomal DNA result may still give an mtDNA result. Just like Y-chromosomal DNA results can be used to help establish male familial relationships, mtDNA results can be used to help confirm (or disprove) a relationship in the female lineage, as mtDNA is transmitted unchanged from a female parent to all (male and female) children. It is common practice in forensic genetics to determine the rarity of a mtDNA haplotype by searching the profile in question in dedicated mtDNA haplotype databases.

The largest and highest quality, freely available mtDNA database is the EDNAP Mitochondrial DNA Population Database EMPOP (<http://empop.online>), which offers the following features:

- a) EMPOP offers tools and help for quality control of population datasets and individual sequences deriving from evidentiary samples. EMPOP is conducting quality control in scientific studies on mtDNA for the leading forensic genetic journals as a requirement before manuscript submission actually takes place.
- b) EMPOP uses alignment-free haplotype searches to guarantee that matches are found in the database, regardless of the alignment used.
- c) EMPOP v4/R12 is designed to offer haplogroup determination of mtDNA sequences based on a maximum likelihood concept.

Additionally, mtDNA information is included in DNA databases as differences between the investigated DNA sequence and the Revised Cambridge Reference Sequence (rCRS). As with Y-chromosomal markers, searching is also possible but as this implies a familial search, special permission from the competent authorities may be required. Depending on local legislation, storing such data may not be permissible. Because differences between a mtDNA sequence or haplotype and the rCRS can sometimes be labelled in different ways, clear rules to indicate these differences should be implemented to avoid false exclusions. A better alternative could be a sequence-based comparison<sup>30</sup>.

**ENFSI recommendation 16** Databases may contain autosomal STR profiles only. For those databases containing profiles from non-autosomal STR profiles or mitochondrial DNA sequences, specific operating

<sup>28</sup> Nothnagel M, et al., (2012) Collaborative genetic mapping of 12 forensic short tandem repeat (STR) loci on the human X chromosome. *Forensic Sci Int Genet.* 6: 778-84

<sup>29</sup> For more information about amelogenin anomalies see: <http://www.cstl.nist.gov/biotech/strbase/Amelogenin.htm>

<sup>30</sup> Parson W, Gusmão L, Hares DR, Irwin JA, Mayr WR, Morling N, Pokorak E, Prinz M, Salas A, Schneider PM, Parsons TJ (2014) DNA Commission of the International Society for Forensic Genetics: revised and extended guidelines for mitochondrial DNA typing. *Forensic Sci Int Genet.* 13: 134-42

Just RS, et al. (2015) Full mtGenome reference data: development and characterization of 588 forensic-quality haplotypes representing three U.S. populations. *Forensic Sci Int Genet.* 14: 141-55.

procedures must be in place to avoid unintended familial searches. To avoid false exclusions, clear rules should be in place to indicate differences between a mtDNA sequence and the rCRS when comparing mtDNA results.

### **3.13 Universal DNA database**

From time to time, politicians initiate discussions regarding the establishment of a DNA database for all inhabitants (and visitors) of a country. The reasoning behind this is to solve more crimes and identify more unidentified human remains. Several years ago, there were plans in the United Arab Emirates to do this and, more recently, Kuwait announced a law to make this possible<sup>31</sup>. In Europe, however, this is not very likely to happen, as it violates Article 8 of the European Convention on Human Rights. In the United Kingdom, about 1.7 million DNA profiles were removed from the national DNA database, because a verdict from the European Court on Human Rights determined that their unlimited storage was in conflict with this article.

Most recently, in the USA, the idea of a universal DNA database has been raised in conjunction with the growing popularity (and therefore utility to law enforcement authorities) of commercial DNA databases<sup>32</sup>. Even in the context of specific crimes, there are many arguments regarding privacy and consent against the creation of such a database<sup>33</sup>.

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<sup>31</sup> <http://news.kuwaittimes.net/website/kuwait-to-enforce-dna-testing-law-on-citizens-expats-visitors-tests-wont-be-used-to-determine-genealogy-affect-freedoms/>

<sup>32</sup> <https://www.genengnews.com/news/universal-dna-database-could-keep-police-investigations-in-bounds/>

<sup>33</sup> <https://www.nextgov.com/ideas/2019/02/dangers-mandatory-dna-database/155028/>

## 4 Deletion criteria

In this chapter, the reasons to remove a DNA profile from the DNA database are discussed. Regardless of the reason for deletion, the removal of a DNA profile should always be recorded in a verifiable way, including the reason for deletion. Deleting a DNA profile from the DNA database may also require the destruction of the cell material as well as hard copies of the DNA profiles and their electropherograms. Deletion of DNA profiles from back-ups or analytical data files is usually more difficult to do.

### 4.1 *End of maximum storage time*

In most countries, there is a maximum time during which DNA profiles are stored. Below is a list of criteria used by different countries for reference samples:

- Fixed time after inclusion
- Variable time after inclusion, depending on the type of crime
- Variable time after inclusion, depending on repeated convictions
- Until the death of a person
- Fixed time after the death of a person
- Variable time after the death of a person, depending on the type of crime
- Fixed time after the completion of sentence
- Variable time after the completion of sentence, depending on the type of sentence or sentence history
- Until no longer relevant (criterion from data protection legislation)

In all but the first two situations, the custodian of the DNA database is dependent on external information to determine the deletion date of a DNA profile. In these cases, the custodian should have access to this information, preferably by means of automated messages, delivered following the event which influences the deletion date of a DNA profile.

**ENFSI recommendation 17** If the removal of a DNA profile from the DNA database is dependent on external instruction from an authorized agent, a process should be in place to inform the custodian of the DNA database of this instruction, preferably by means of an automated message.

For non-matching DNA profiles of stains, the storage time is fixed or variable, depending on the type of crime or the statute of limitation for the crime. For non-matching DNA profiles of stains, see § 4.3.

### 4.2 *Non-conviction of a person*

Suspects, arrestees and convicted persons who have successfully appealed their conviction may have to be removed from the DNA database. If this is prescribed by law, the custodian of the DNA database must receive or have access to information regarding convictions or acquittals of any persons included in the DNA database. Experience in several countries has shown that this kind of information is not always provided in time by the courts or the public prosecution service. This has resulted in matches with persons who should have been removed from the DNA database, and courts have ruled that these matches are inadmissible as evidence. The ENFSI recommendation in the previous paragraph is equally applicable to this removal condition.

### 4.3 *Match of stain with person*

When a reference DNA profile matches a DNA profile from a crime scene stain in the DNA database and the match has been processed by the judicial authorities, the latter may be removed from the DNA database because it has fulfilled its purpose. If the match occurs within the same case, this is called a “benchmark match”. In some countries (like the Netherlands), a crime scene DNA profile cannot be removed from the DNA database until the custodian of the DNA database has received a message that either the suspect has been convicted, or that the prosecution has decided not to use DNA evidence. The ENFSI recommendation in paragraph 4.1 is equally applicable to this removal condition. For various reasons, countries may retain crime scene stain profiles in their DNA database even after they have shown

a match with a person. The Nuffield Council for Bioethics even recommended this in their 2007 Bioethics report, to verify possible future doubts about a match<sup>34</sup>.

#### **4.4 Duplication**

Persons may or may not be sampled repeatedly for inclusion in the DNA database, depending on the legislation of the country, although sometimes this may also occur inadvertently. An inadvertent duplicate is a waste of resources, therefore a system that can be consulted by those responsible for sampling should be implemented, through which they can verify whether a person is already present in the DNA database.

##### **ENFSI recommendation 18**

There should be a system that can be consulted by those responsible for taking reference samples, to verify whether a person is already present in the DNA database.

Sometimes people use a false identity, and for this reason, duplication of sampling is not always avoidable. Therefore, a rapid biometric identification system like fingerprints should be linked to the system, indicating whether a person is already present in the DNA database.

The analysis of unintentional and (low level) intentional duplicates, however, is a useful quality control instrument. When removing a duplicate, the sample with the least chance of being removed in the future should be kept (if legally possible). Duplicates produced with partially non-overlapping sets of loci are, of course, also useful to keep (e.g. Powerplex 16 and Identifiler).

#### **4.5 Match with elimination database**

Any DNA database should have an associated so-called elimination DNA database (or databases), containing the DNA profiles of persons who could introduce cross-contamination to the investigated traces. Such elimination databases should include anybody handling the DNA samples in the DNA lab, as well as those cleaning the labs or performing any other kind of maintenance. Also, people from earlier in the chain of custody, such as investigating officers and other persons present at the crime scene, should be included. In addition, unidentified DNA profiles found in negative controls, which may come from people involved in the manufacturing of disposables and/or chemicals, should be included and shared with other ENFSI countries.

When a DNA profile in the DNA database matches a DNA profile from the elimination DNA database, it should be deleted because it is not meant to be included. However, this should not be done before the contamination incident has been analyzed thoroughly, the presumed cause of the match (contamination) has been confirmed, and actions to prevent this (and similar) accidents occurring in the future have been formulated. Laboratories supplying DNA profiles to the DNA database may have their own elimination databases to exclude their own employees as a possible source of contamination. In most countries, there is no specific legal basis for the establishment of an elimination database. However, because personal data are involved, laboratories, which, for quality control reasons, have decided to establish an elimination database, are bound by the data protection law of their country. These laws usually require the explicit written consent of the persons to be included into the elimination DNA database. In addition, employers may include willingness to be included in the elimination database as a job requirement.

**ENFSI recommendation 19** DNA databases should contain an associated elimination DNA database (or databases). This should include laboratory staff of all categories, as well as visitors and maintenance personnel and profiles from those with access to traces (e.g. police, crime scene technicians).

<sup>34</sup> <http://nuffieldbioethics.org/project/bioinformation/>

Manufacturers of disposables and/or chemicals should follow the joint recommendations of ENFSI, SWGDAM and SMANZL<sup>35</sup>, which have recently been converted into the ISO18385 standard, to prevent the contamination of their products.

ICMP<sup>36</sup> has developed a manufacturers' elimination database (MED)<sup>37</sup>, which was devised in concert with the forensic DNA community, based on ICMP's independent status and data protection capabilities. The application has been successfully tested and launched in 2018. It is intended to provide forensic DNA laboratories with the ability to query a database of DNA profiles of individuals, acquired from the staff of participating companies involved in the forensic DNA supply chain, to avoid the inadvertent inclusion of manufacturers' staff profiles in either forensic DNA databases or investigations. Countries wishing to upload profiles can email **tmpusr@icmp.int** to set up an account.

**ENFSI recommendation 20** Because elimination databases are not shared with other EU/ENFSI countries, unidentified DNA profiles found in negative controls, which may originate during the manufacture of disposables and/or chemicals should be uploaded to the ICMP Manufacturers Exclusion Database, MED

#### **4.6 *New information demonstrating that the DNA profile should not have been included***

Occasionally, during a police investigation, new information becomes available showing that a trace, which was thought to be relevant to the crime, has an origin that is not relevant to the crime. Additionally, a person may accidentally have been asked or ordered to give a buccal swab related to a crime illegally (for reasons not permitted by law). If such a DNA profile has already been included in the DNA database, it must be removed as soon as possible to prevent the presence of unauthorized DNA profiles in the DNA database.

**ENFSI recommendation 21**

Policies and procedures should be in place to ensure that DNA profiles deemed no longer relevant by the authorizing agent are deleted.

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<sup>35</sup> Manufacturer contamination of disposable plastic-ware and other reagents—An agreed position statement by ENFSI, SWGDAM and BSAG. Forensic Science International: Genetics, Volume 4, Issue 4, July 2010, Pages 269-270.

<sup>36</sup> See paragraph 23.7.1 for more information about ICMP

<sup>37</sup> <http://www.icmp.int>

## 5 Matching rules

This chapter describes the criteria which are used to determine whether two similar DNA profiles are a match.

### 5.1 Match/hit definition

The words “match” and “hit” are sometimes used in different ways. The Dutch police use the word “match” if the DNA profiles of crime-related stains are similar, and the word “hit” if the DNA profile from a crime-related stain is similar to the DNA profile of a reference sample. In the USA, the word “match” is used if two DNA profiles in the CODIS DNA database correspond to each other, and the word “hit” is used if a match is confirmed by a DNA expert. In this document, we use the ENFSI definition<sup>38</sup>, which does not differentiate between a hit and a match:

*Hit/Match: A confirmed match between DNA profiles discovered by a database search at a single instant in time. It can be stain to stain or stain to person.*

In this document, the word ‘match’ will be used.

### 5.2 Search modes

DNA profiles can be compared in different ways. In CODIS, these are called search stringencies:

- “High stringency” means that all alleles of every locus present in one DNA profile must also be present in the matching DNA profile in exactly the same amount;
- “Moderate stringency” means that, of two DNA profiles, the alleles of a locus with the least number of alleles must be present in the corresponding locus of the other DNA profile. This stringency is used when comparing mixed DNA profiles with single DNA profiles. Because homozygotes are designated by a single allele value in CODIS, searching at moderate stringency with single DNA profiles also detects an allele drop-out in one of the DNA profiles compared (e.g. 12/13 will also match the apparent homozygotes 12/ or 13/);
- “Low stringency” means that, in each locus compared between two DNA profiles, at least one allele of that locus must be present in the other DNA profile. This stringency is used to find parent-child relationships.

Table 2 provides some examples of match results when a target<sup>39</sup> profile (15,16) is searched against different candidate profiles using the basic CODIS stringencies.

Target profile	15,16	Match stringency		
		High	Moderate	Low
Candidate profile	15,16	Match	Match	Match
	15 <sup>40</sup>	No match	Match	Match
	15,17	No match	No match	Match
	17,18	No match	No match	No match
	15,16,17	No match	Match	Match
	15,16,17,18	No match	Match	Match

*Table 2: Examples of match results when a target profile (15,16) is searched against different candidate profiles in CODIS*

<sup>38</sup> [http://www.enfsi.eu/sites/default/files/documents/enfsi\\_dna\\_wg\\_terms\\_and\\_abbreviations\\_0.pdf](http://www.enfsi.eu/sites/default/files/documents/enfsi_dna_wg_terms_and_abbreviations_0.pdf)

<sup>39</sup> In CODIS, the profile with which the search is conducted, is called the target profile

<sup>40</sup> In CODIS a homozygote is designated with a single allele value.

In CODIS, mixed profiles cannot be compared to each other to find common donors. Recently, however, a program was developed which can be used to find common donors in mixed profiles exported from CODIS<sup>41</sup>.

In some countries, a search strategy called “familial searching” is allowed. This means that, apart from searching for full matches, a search for matches with possible relatives of the donor of a crime scene associated DNA profile can also be conducted.

This search strategy may use the above-mentioned “low stringency” search mode to find possible parent-child relationships but may also search for profiles which:

1. share higher than the average number of alleles in random unrelated DNA profiles (which may indicate a possible sibling),
2. contain rare alleles (which may indicate a possible family member),
3. have a high likelihood ratio and therefore provide, for example, very strong support for the proposition that the persons are related (e.g. are siblings) rather than for the proposition that the persons are unrelated.

From a statistical point of view, the 3<sup>rd</sup> approach is the preferred strategy<sup>42</sup>. The programs mentioned in §23.6 may be used to perform a familial search against a national DNA database. This requires, however, that all DNA profiles of persons from a national DNA database are present in or can be exported to this program. The outcome of the search is a starting point to find the true owner of the crime scene stain via tactical police work. This police work may be preceded or accompanied by Y-chromosomal and/or mitochondrial DNA testing, to decrease the number of candidates and/or their priority order. Simulation studies have been published showing the number of candidates a familial search in a DNA database may yield<sup>43,44,45,46</sup>.

The outcome of the search may point in the wrong direction, in the same way that a match may turn out to be an adventitious match. The search results should therefore be reported with a warning similar to the warning mentioned in Recommendations 25 and 26. An extensive review of the ethical aspects of familial searching has been written by Professor Sonia Suter<sup>47</sup>.

In recent years a growing trend has emerged where commercially available genealogy databases have been used by investigating police in the USA to identify relatives of suspects in crime. However, this practice raises many ethical issues surrounding data protection, privacy and consent, since most people do not expect to participate in a criminal investigation when submitting their biological sample to commercial databases<sup>48,49</sup>. In September of 2019, the US Department of Justice published their guidelines for the use of such databases in criminal investigations<sup>50</sup>: “The policy says “forensic genetic genealogy” should generally be used only for violent crimes such as murder and rape, as well as to identify human remains. (The policy permits broader use if the ancestry database’s policy allows such

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<sup>41</sup> K. Slooten (2017) *Forensic Science International: Genetics* 26,40-47

<sup>42</sup> D.J. Balding et al (2013) *Decision-making in familial database searching: KI alone or not alone?* *Forensic Science International Genetics* 7, 52-54.

<sup>43</sup> Hicks et al. (2010) *Forensic Science International: Genetics* 4 (5), pp. 316-322. Use of DNA profiles for investigation using a simulated national DNA database: Part II. Statistical and ethical considerations on familial searching.

<sup>44</sup> C. van Kooten et al. (2010) Poster nr 9 presented at the 21<sup>st</sup> International Symposium on Human Identification. It’s all relative(s): Familial Searching in the Netherlands. ([http://dnadatabank.forensischinstituut.nl/Images/lr-52634-poster-a0-verwantschapsonderzoek\\_tcm127-464088.pdf](http://dnadatabank.forensischinstituut.nl/Images/lr-52634-poster-a0-verwantschapsonderzoek_tcm127-464088.pdf)).

<sup>45</sup> Ge et al (2011) *Journal of Forensic Sciences* Volume 56, Issue 6, pages 1448–1456, November 2011  
Comparisons of familial DNA database searching strategies.

<sup>46</sup> K. Slooten, and R. Meester (2014). "Probabilistic strategies for familial DNA searching." *Journal of the Royal Statistical Society: Series C (Applied Statistics)* Volume 63, Issue 3, pages 361–384, April 2014

<sup>47</sup> <http://jolt.law.harvard.edu/articles/pdf/v23.2/23HarvJLTech309.pdf>

<sup>48</sup> <https://www.theatlantic.com/science/archive/2019/10/genetic-genealogy-dna-database-criminal-investigations/599005/>

<sup>49</sup> <https://www.sciencenews.org/article/forensic-genetic-genealogy-companies-police-privacy>

<sup>50</sup> <https://www.sciencemag.org/news/2019/09/new-federal-rules-limit-police-searches-family-tree-dna-databases>

searches.<sup>51</sup>) Police should first exhaust traditional crime solving methods, including searching their own criminal DNA databases”<sup>52</sup>.

### 5.3 Number of matching loci/match probability

The number of matching loci depends on the number of loci present in the DNA profiles to be compared. The lower the number of loci, the higher the match probability of the DNA profile, and the higher the chance of an adventitious match, especially with large DNA databases. For this reason, DNA profiles included in the DNA database on a permanent basis should have a minimum number of loci, or, better yet, a maximum random match probability, as indicated in § 3.3. For reference samples, the number of loci is usually 10 or more, to increase the chance of finding a match with a (partial) DNA profile of crime-related biological material. At a national level, a lower number is also possible, but in this case, the DNA profile should have a low match probability. This is the case in Germany, which, until a few years ago, used the 7 old ESS loci, plus the highly discriminating locus SE33. The matching rules of the EU-Prüm implementation decision require a minimum number of 6 fully matching loci.

### 5.4 Near matches

Several situations may lead to a near match (where one locus does not match or does not match completely) between two DNA profiles of the same person:

- A human error made during the production of one of the profiles. This may, for instance, happen when an allele is incorrectly called, a mixed profile is incorrectly split up into one or more of its contributors, or a typographical error is made while the DNA profile is entered manually into the DNA database. When setting up a new DNA database, the allele calling and the DNA database import process should be automated as much as possible to avoid this problem. Manually entering DNA profiles into the DNA database has been shown to be the greatest source of errors, hence this should be conducted using a process that detects typographical errors, such as the double-blind method (entering a DNA profile twice without seeing the first one and the database software checking if both entries are identical).

**ENFSI recommendation 22** The occurrence of errors in DNA profiles as a result of human mistakes associated with data entry should be avoided as much as possible by automating the allele calling and the DNA database import process. Automated processes reduce the possibility of human error, however, when DNA profiles are entered manually into the DNA database, a process that detects typing errors, for example the double-blind method of entry, should be used.

- An allele drop-in or drop-out due to low-level DNA profiling of one of the DNA profiles (see §3.5)
- The occurrence of so-called “null alleles”. These are alleles which are not amplified during the PCR reaction due to a mutation in the primer binding-site region. When two STR-typing kits use different primers for the same heterozygous locus, and the DNA of a person contains a mutation in the primer binding region used in one kit but does not contain a mutation in the primer region used in the other kit, the former kit will detect only one allele (apparent homozygote) and the latter will detect two alleles (heterozygote). The presence of a null allele may be detected by the unexpected low peak height of the apparent homozygote, but this requires an attentive DNA analyst or intelligent allele-

<sup>51</sup> Initially, the US police used the database of GEDmatch secretly. When GEDmatch found out about this unexpected use of its database, it told its customers that their DNA data could also be used only for the investigation of rape and murder cases and advised people to opt-out if they did not want their data to be used for this purpose. However, when the police uploaded a DNA profile from a case which did not fulfil these criteria, it reversed its policy and opted everybody out and required an active opt-in from people to allow the police to use their DNA data to identify relatives of suspects. Very recently, however, the Florida police obtained a warrant to search the whole GEDmatch database, including all those who had opted out (<https://futurism.com/cops-warrant-entire-dna-websites>). 23andMe, another DNA genealogy company, has indicated that if they were to receive such a warrant, they would use every legal remedy possible to challenge it (<https://blog.23andme.com/news/our-stance-on-protecting-customers-data/>).

<sup>52</sup> <https://www.sciencemag.org/news/2019/09/new-federal-rules-limit-police-searches-family-tree-dna-databases>



calling software<sup>53</sup>. A special case of a null allele, not caused by a primer binding-site mutation but by a deletion instead, is the disappearance of the Y-amelogenin allele, which makes the DNA profile of a male look female.

- The occurrence of shifted allele size in one of the DNA profiles. When 2 kits use different primers for the same heterozygous locus and the DNA of a person contains a deletion or an insertion after the primer region used in one kit but before the primer region used in the other kit, there will be a shift in the size of an allele in one DNA profile compared to the other (e.g. 10.3 and 11).

Given the above-mentioned phenomena, only searching a DNA database for full matches (high stringency) may lead to missed matches (false negative matches). To find false negative matches, a “less stringent” search strategy must be used, either permanently or occasionally. Countries such as Switzerland, the Netherlands and the United Kingdom already perform this kind of quality control check regularly, by searching for near matches, which are then checked for possible errors. Searching for near matches may lead to matches with close relatives, hence the pros and cons of this strategy should be evaluated in advance (see also chapter 9). The software used for the international comparison of DNA profiles under the terms of the EU Prüm decision also allows for one mismatch, to detect near matches<sup>54</sup>. After finding such a match, both countries must contact each other to verify the original data and the processing methods. Near matches involving 8 loci or less often prove to be adventitious (false positive), but it may be worth investigating further, if this can assist in the investigation of a serious case (see also chapter 9).

The above-mentioned stringencies in CODIS can be adapted and combined to create more robust searches that can take into account allele drop-in and drop-out in a more flexible manner, thus increasing the chances of a match being returned. For example:

- A “Partial loci” search can be initiated by marking all loci to High stringency, with the exception of those loci marked as partial when inputting the data – these should be set to Moderate stringency. This setting guarantees that all known (complete) loci will be evaluated as such, but will find more potential matches for the partial loci. Matches otherwise missed will be included in search results and can be reviewed accordingly.
- The “Floating moderate” search can be initiated by setting all loci to High stringency, while configuring the search to allow for a specific number of loci to match at Moderate stringency.

Other dedicated search strategies can be developed and adapted for different situations to return the best possible results, which can then be reviewed accordingly to determine if they are true, investigatively useful matches or not.

**ENFSI recommendation 23** To prevent and detect false exclusions (e.g. true matches that are not found due to an error in one of the DNA profiles), DNA profiles should be searched using a full Database search allowing at least one mismatch or other dedicated search strategy. The original data of DNA profiles involved in such near matches should be checked for possible errors during their production and processing.

## 5.5 Match nomenclature

A specific nomenclature for matches was developed by members of the DNA database and DNA legislation subgroup, the ENFSI DNA Working Group. It consists of 2 figures and a letter. The first figure indicates the number of fully matching loci, the second figure indicates the number of mismatches and the letter indicates the match stringency: H(igh), M(oderate) or L(ow). So a 14-1-H-match means a high stringency 14 locus match plus 1 mismatch.

<sup>53</sup> More information about the occurrence of null alleles can be found at: <http://www.cstl.nist.gov/biotech/strbase/NullAlleles.htm>.

<sup>54</sup> Forensic DNA Profiles Crossing Borders in Europe (Implementation of the Treaty of Prüm). Profiles in DNA 2011 (<http://www.promega.com/resources/articles/profiles-in-dna/2011/forensic-dna-profiles-crossing-borders-in-europe/>).

## 5.6 Match validation

There are several reasons why a DNA database match may need to be validated:

- Confirmation of the original DNA analysis:
  - Some countries require a new sample to be taken from the suspect and have that new sample re-analyzed.
  - Some countries perform a second analysis on a duplicate sample previously taken from the involved person but not yet analyzed.
  - Some countries require a new sample and re-analysis, because a database match may influence a jury in court (because this is an indication of earlier convictions).
  - Some countries do an independent duplicate analysis for all their reference samples, avoiding any match validation needs.
  - The requirement for a duplicate analysis may be linked to a suspect making a plea of not guilty, and contesting the DNA evidence.
- Possibility of an adventitious match:  
In this case, more loci should be analyzed to decrease the possibility of an adventitious match.
- Near match (one allele does not match):  
In this case, the original data of both DNA profiles should be checked to eliminate the possibility of a typing or allele calling error.
- Match with a mixed DNA profile:  
A DNA database match based on numbers of a single DNA profile with a mixed DNA profile is not necessarily a true match (see § 3.8). A DNA expert should indicate whether this type of match can be a true match or not.

## 6 Dispositioning

After finding a candidate match in the DNA database, this match must be confirmed. When a match is found between two full DNA profiles, this confirmation can be done by specially qualified DNA database personnel or using an automated process. However, matches with partial and/or mixed profiles must be examined and assigned a final disposition by a DNA expert. Usually, the final disposition of a match can also be registered in the DNA database to prevent the same match from being reported again after a new search action.

### 6.1 Match counting

One of the parameters to determine the efficiency of a DNA database is the number of matches it generates. Counting the matches between two DNA profiles is easy. In serial crimes committed over a period of time, however, different approaches are possible. Table 3 shows the number of matches that will be found when a single (unknown) individual commits a series of 7 crimes over time and leaves DNA at each crime scene.

DNA profile	Nr of matches	Description of the matches
A	-	
B	1	B -> A
C	2	C -> A&B
D	3	D -> A&B&C
E	4	E -> A&B&C&D
F	5	F -> A&B&C&D&E
G	6	G -> A&B&C&D&E&F
H	7	H -> A&B&C&D&E&F&G
Total	28	

Table 3. Number of matches that will be found when a single (unknown) individual commits a series of 7 crimes over time and leaves DNA at all 7 crime scenes.

For a series of X crimes, the number of matches is  $(X-1)X/2$ . For high volume crime cases, this manner of counting leads to match counts which are not representative when compared to the number of cases

involved. This is why the ENFSI counts matches in serial crimes in a different manner. The following definition is taken from the document “ENFSI DNA Working Group Terms and Abbreviations”<sup>55</sup>:

*For statistical purposes, matches with multiple identical profiles from the same case will be counted as one match, but as separate matches if they originate from different cases. In serial crimes, the total number of matches is N-1 to the number of matching profiles (e.g. a series of 8 identical stain profiles from different crimes yields 7 stain-to-stain matches). If subsequently the DNA profile of a person matches the series, it yields 8 stain-to-person matches. The number of stain-to-stain matches should then be removed from statistics.*

An expression that is also used in match counting is “the number of investigations aided”. This equals the number of DNA profiles involved in matches. In the example above, dealing with a series of 8 identical DNA profiles, there are 7 matches and 8 investigations aided.

A series of matching DNA profiles may be given a unique identification code to indicate that they are similar. In the Netherlands, this is called the DNA cluster-number, which has proved to be very useful for investigators in designating the series.

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<sup>55</sup> [http://www.enfsi.eu/sites/default/files/documents/enfsi\\_dna\\_wg\\_terms\\_and\\_abbreviations\\_0.pdf](http://www.enfsi.eu/sites/default/files/documents/enfsi_dna_wg_terms_and_abbreviations_0.pdf)

## 6.2 Output/efficiency measurement

The output of a DNA database is the number of matches it generates. Simon Walsh et al.<sup>56</sup> have published a formula which describes the output of a DNA database:

$$H = \frac{\alpha N}{M} \times \omega C$$

Where...

*H = number of hits/matches*

*N = number of persons in the 'offender' database*

*M = active criminal population*

*C = number of crimes in the 'forensic' database*

*α = quality factor (person sampling)*

*ω = quality factor (crime/exhibit sampling)*

The two quality parameters in the formula determine the efficiency of a DNA database. If H, N, M and C are known, the product of the two quality factors can be determined by transforming their formula into:  $\alpha\omega = HM/NC$ .

Walsh et al. propose an efficiency measurement parameter, the return index (RI), where  $RI = H/NC$ . As this parameter is inversely proportional to the size of the database, it wrongly suggests that large DNA databases are less efficient than smaller DNA databases. The ENFSI DNA Working Group proposes the use of two different DNA database performance parameters, which express two different types of efficiencies:

H/C: the number of stain-to-person matches, relative to the number of stains included in the DNA database (also known as the match rate). This parameter expresses the chance that a stain profile included in the DNA database will match a reference profile. This is a very important parameter, because it shows the crime-solving capacity of the DNA database and whether the right items were collected by the crime scene officers. It is self-evident that more stains will be matched to a person as more members of the (criminal) population are included in the DNA database. So, as the size of the DNA database increases, H/C will increase. One can determine the match rate at a certain point in time (including all stains that have ever been included) or during a certain time interval. The UK for instance determines the match rate per month so they measure how many matches were found with the stains that were uploaded in a certain month. The first approach also includes matches which were found after stains that initially did not match a person, match a person that was included at a later date.

H/N: the number of stain-to-person matches relative to the number of persons included in the DNA database (the percentage of persons in the database that have caused matches). This parameter indicates whether the right people have been sampled for inclusion in the DNA database. This parameter is also important, because it does not make sense to allocate resources for including people into the DNA database who will never cause matches (even though it is a one-time process). To properly calculate this parameter, duplicate person profiles and additional matches with the same person should be excluded.

Also this parameter can be followed over time, and it can also be applied to subgroups of persons in the DNA database. In the Netherlands, for example, this ratio was 0.52 for suspects (in 2005) and 0.06 for convicted persons (in 2006).

Because the policies for keeping or removing DNA profiles from stains and persons in a DNA database are different in different countries, table 4 cannot be used to compare the DNA database performance of the different ENFSI countries using the parameters H/C and H/N.

The number of stain-to-stain matches can either be expressed as the number (or percentage) of stains involved in matches (investigations aided), or as the number (or percentage) of profiles giving a match at

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<sup>56</sup> S.J. Walsh et al (2010) Modeling Forensic DNA Database Performance; J. For. Sci. 55(5) 1174-1183

inclusion, which is lower, because the first profile of a cluster does not result in a match (see Table 2 in § 5.7).

Country	Population size	Persons					Remarks	Prum	Stains		Matches				Date	Stain-person matches per person	
		A	S	CO	T	Prum			Remarks	Person/Stain	Stain/Stain	Total	Remarks				
Albania	3,600,000																
Armenia	3,000,000																
Austria	8,800,000				230,359	230,359	137,848	40,760	Other:specify		29,729	13,495	40,002	Other:specify	Dec-18		
Belgium	11,413,203		2,185	50,217	52,382	52,382	53,992	29,962							Dec-18		
Bosnia & Herzegovina	4,400,000								Including duplicates								
Bulgaria	7,900,000																
Croatia	4,300,000				31,199			5,320					6,452		Apr-14		
Cyprus	772,000		379		379		15,651	3,832	No duplicates		219	93	312	139	451	Cold and warm hits	
Czech Republic	10,638,000				226,802	226,760	21,756	20,290	Persons added minus persons removed				22,903	10,242	33,145	Cold and warm hits	
Denmark	5,700,000				130,662	130,662		19,957	Including duplicates				34,813	5,585	40,398	Only cold hits	
Estonia	1,318,700				52,697	33,933		10,265	Persons added minus persons removed				6,698	1,201	7,899	Only cold hits	
Finland	5,509,242				180,591	180,591		12,984	Persons added minus persons removed				31,243			Only cold hits	
France	66,992,699	154,227	3,589,381	577,751	4,167,132	4,167,132		542,866	No duplicates				240,781	49,592	290,373	Cold and warm hits	
Georgia	4,700,000		6,962	44	11,570			7,320	Including duplicates				35	19	54	Only cold hits	
Germany	83,000,000				871,506	852,369		320,719	Persons added minus persons removed				205,270	55,206	260,476	Dec-17	
Greece	10,600,000				14,419			17,779	Persons added minus persons removed				1,651	1,971	3,622	Dec-18	
Hungary	9,982,000				202,471	141,246		9,581	Including duplicates				3,375	383	3,758	Cold and warm hits	
Iceland	315,000								Persons added minus persons removed								
Ireland	4,200,000		12,409	3,103	15,512			4,042	Including duplicates		1,016	254	1,270	108	1,378	Only cold hits	
Italy	60,589,445			4,800		4,800		11,000	No duplicates				50	450	500	Only cold hits	
Kosovo	1,800,000																
Latvia	1,900,000		50,145	10,370	60,515	60,515		11,140	Including duplicates				3,026	373	3,399	Jun-17	
Liechtenstein	37,000				371	0		341	No duplicates				284	82	366	Dec-17	
Lithuania	2,960,000				92,463	92,463		6,168	Including duplicates				3,348	521	3,869	Jun-17	
Luxembourg	602,000		582	2,790	3,372	3,322		7,048	Persons added minus persons removed		312	339	651	22,187	22,838	Cold and warm hits	
North Macedonia	2,100,000				22,998			10,473	All person profiles ever added				2,698	323	3,021	Only cold hits	
Malta	400,000		30		30			430					1	3	4	Jun-15	
Montenegro	650,000																
Netherlands	17,000,000				267,407	267,407		70,460	Persons added minus persons removed				60,544	5,464	66,008	Cold and warm hits	
Northern Ireland	1,685,000								No duplicates								
Norway	5,300,000		6,286	89,909	96,195			1,263	No duplicates		4,550	12,501	17,051	2,061	19,112	Only cold hits	
Poland	38,200,000				74,841	74,841		11,045	No duplicates				1,519	1,141	2,660	Only cold hits	
Portugal	10,300,000		7,430	7,587	7,398	Other: specify	2,409	2,346	Prüm Stains category includes also Missing Persons, Unidentified Human Remains and Unidentified Person's profiles		134	73	207	63	270	Cold and warm hits	
Romania	22,000,000		2,612	37,401	40,013	40,013		1,501	Persons added minus persons removed		174	1,073	1,247	597	1,844	Cold and warm hits	
Russia	143,800,000																
Scotland	5,500,000		174,219	136,888	311,107			18,725					31,249	2,556	33,805	Apr-13	
Serbia	7,335,000				68,643			12,674					7,018	2,372	9,390	Dec-18	
Slovakia	5,500,000				34,426	34,273		7,677	Including duplicates				5,915	1,185	6,687	Only cold hits	
Slovenia	2,000,000								Persons added minus persons removed							National and Prüm hits	
Spain	46,570,000				366,562	364,961		110,296	Persons added minus persons removed				59,556	53,704	113,260	Cold and warm hits	
Sweden	10,230,185		12,439	150,258	162,697	162,697		36,776	Including duplicates				59,453	18,625	78,078	Cold and warm hits	
Switzerland	7,779,000				193,651	0		82,857	Persons added minus persons removed		31,463	27,990	59,453	18,625	78,078	Cold and warm hits	
Turkey	66,800,000								No duplicates				71,618	18,293	89,911	Cold and warm hits	
UK (England & Wales)	53,700,000				5,018,728			597,029					2,029,892	427,287	2,457,179	mt/19	
Ukraine	47,600,000																
Total	808,159,774				13,009,287			2,225,086					2,933,407	695,228	3,600,209		0.23

A = Arrestees S = Suspects CO = Convicted offenders T = Totals (or when no distinction can be made)

When using these data one should realize that:  
# There are countries that add more than one DNA-profile of a person to their DNA-database  
# There are countries that add more than one unique DNA-profile of a stain per crime scene to their DNA-database  
# Counties use different removal regimes for DNA-profiles of stains: never or immediately after a match with a person or after the authorities have dealt with the match with the person  
# Counties use different removal regimes for DNA-profiles of persons: After some storage time and/or if a person is not prosecuted or convicted  
# Stain-to-person matches can be so-called "cold hits" (matches for which no suspect was known) or matches where both a stain and a suspect were added to the DNA-database  
# One person can match more than one stain

Table 4. Annual ENFSI DNA database overview for 2018.

As a national DNA database is regularly subject to attention from the public, politicians and the media, a DNA database manager should consider establishing performance parameters and making these publicly available.

### ENFSI recommendation 24

As a national DNA database is regularly subject to attention from the public, politicians and the media, a DNA database manager should consider establishing tools to monitor the effectiveness of their DNA database and communicating this objective information publicly.

The above-mentioned performance parameters only apply to the performance of the DNA database itself. The efficiency of a DNA database as a tool to investigate and solve crimes also depends on many other factors, which have been reviewed by Bieber<sup>57</sup>. The fate of 625 international matches reported to the

<sup>57</sup> F.R. Bieber (2006) Turning base hits into earned runs: Improving the effectiveness of forensic databank programs. Journal of Law, Medicine & Ethics 34(2) p 222-233.

Dutch authorities in 2010 was investigated during the ISEC-PIES project<sup>58</sup>. Only 37 of those matches were used in court. Jennifer Doleac has investigated the effects of DNA databases on crime in the USA<sup>59</sup>. Amankwaa and McCartney have examined the effectiveness of the UK national DNA database from a more financial point of view<sup>60</sup>. Additionally, the efficiency of DNA databases in the context of cross-border exchange of data has also been examined within the scope of the EU Prüm Decisions<sup>61</sup>.

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<sup>58</sup> [http://dnadatabank.forensischinstituut.nl/Images/report-pies-t5-taverne-broeders-opmaak-ul-26112015-isbn-def-met-omslag\\_tcm127-612193.pdf](http://dnadatabank.forensischinstituut.nl/Images/report-pies-t5-taverne-broeders-opmaak-ul-26112015-isbn-def-met-omslag_tcm127-612193.pdf)

<sup>59</sup> Doleac, Jennifer (2016) The effect of DNA databases on crime. *American Economic Journal; Applied Economics* 9(1): 165-201

<sup>60</sup> Amankwaa, Aaron Opoku and Carole McCartney (2019) The effectiveness of the UK national DNA database. *Forensic Science International: Synergy*, Vol.1, 45 – 55.

<sup>61</sup> [http://www.europarl.europa.eu/RegData/etudes/STUD/2018/604971/IPOL\\_STU\(2018\)604971\\_EN.pdf](http://www.europarl.europa.eu/RegData/etudes/STUD/2018/604971/IPOL_STU(2018)604971_EN.pdf)

## 7 Adventitious matches

As DNA databases become larger, the chance of adventitious matches occurring also increases, especially with partial and mixed profiles and the DNA profiles of relatives, which have higher random match probabilities. If a crime stain DNA profile has a random match probability of 1 in 1 million, and a DNA database contains 3 million DNA profiles, a mean of three matches can be expected, and none of them may be the actual originator of the crime stain DNA profile. Therefore, every DNA database manager should be able to determine the chance of finding adventitious matches in their DNA database. Table 5 may be helpful in this respect. This table gives the expected number of adventitious matches when a DNA database of a given size is searched using a DNA profile with a given match probability.

		Size of the DNA database			
		10.000	100.000	1.000.000	10.000.000
Random Match Probability (1:X)	10.000	1	10	100	1.000
	100.000	0,1	1	10	100
	1.000.000	0,01	0,1	1	10
	10.000.000	0,001	0,01	0,1	1
	100.000.000	0,0001	0,001	0,01	0,1
	1.000.000.000	0,00001	0,0001	0,001	0,01
	10.000.000.000	0,000001	0,00001	0,0001	0,001

Table 5: Expected number of adventitious matches when searching a DNA database of a given size using a DNA profile with a given random match probability

The expected numbers of adventitious matches in Table 4 are the expected numbers for one search using a DNA profile with a given random match probability in a DNA database of a given size. On an annual basis, the number of searches is usually much higher than one. Hence, on an annual basis, the expected number of adventitious matches is the expected number of adventitious matches of one search multiplied by the annual number of those searches. So a DNA database in which many crime scene DNA profiles are compared can expect more adventitious matches on an annual basis than a DNA database of similar size in which fewer crime scene DNA profiles are compared per year. An estimation of the annual expected number of adventitious matches can be made by separating the crime-related DNA profiles into match probability classes, and estimating how many of each class are compared to the reference samples in the DNA database.

Table 6 gives a theoretical example of a DNA database containing 4 million reference DNA profiles, with which 70,000 crime-related DNA profiles of different random match probabilities (RMP) are compared on an annual basis, and calculates the expected number of adventitious matches from those figures (but there may be more or less than the expected number).

DNA-database size	RMP crime related stain	Number of searches	Expected Number of Adventitious Matches
4.000.000	1 : 10.000.000.000	50.000	20
	1 : 1.000.000.000	10.000	40
	1 : 100.000.000	5000	200
	1 : 10.000.000	3000	1200
	1 : 1.000.000	2000	8000
Total		70.000	

Table 6: Theoretical example of a DNA database containing 4 million reference DNA profiles, with which 70,000 crime-related DNA profiles of different random match probabilities are compared.

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Another factor that influences the expected number of adventitious matches is the presence of relatives in the DNA database. This results from the fact that the match probabilities between relatives are higher than the random match probability. Table 7 lists the theoretically calculated mean approximate match probabilities between various kinds of relatives, as compared to a random match probability of 1 in 10<sup>12</sup>.

<b>Relationship</b>	<b>Match Probability</b>
No relationship (random match probability)	1 in 10 <sup>12</sup>
First cousin	1 in 10 <sup>11</sup>
Half-sib or uncle/nephew	1 in 10 <sup>10</sup>
Parent or child	1 in 10 <sup>8</sup>
Full-sib	1 in 10 <sup>5</sup>

*Table 7: Approximate match probabilities<sup>62</sup> between various kinds of relatives, as compared to a random match probability of 1 in 10<sup>12</sup>.*

Identical twins, of course, have the same DNA profile.

The exact expected number of adventitious matches due to the presence of relatives in a DNA database is impossible to calculate without knowing the numbers and types of relatives present.

The impact of the presence of relatives in a DNA database on the expected number of adventitious matches seems limited, however, as shown in the next example: If 50,000 full SGM+ DNA profiles from crime-related stains are searched against a DNA database of 4,000,000 reference profiles, and 10% of the crime-related stain donors have a sibling in the DNA database, 5,000 DNA profiles will have a match probability of 1:10,000 instead of 1:10,000,000,000. The extra expected number of adventitious matches caused by the DNA profiles of these 5,000 persons with a sibling in the DNA database is 5,000 x 1/10,000 = 0.5. This is only a small extra number, when compared to the 20 adventitious matches which are expected anyway by searching a DNA database of 4,000,000 reference profiles with 50,000 DNA profiles from crime-related stains of persons who are unrelated. The effect of relatives on the expected number of adventitious matches will increase over time as more persons related to each other in some way will be included in the DNA database. At this moment, we are only dealing with one generation of relatives but in 10 years, a next generation of relatives may also be present. In addition, a recent study of the Danish DNA database indicated that the effect of relatives must not be ignored<sup>63</sup>.

Because the risk of adventitious DNA database matches cannot be neglected, a warning should be included, indicating the factors that increase the possibility of finding an adventitious match (size of the database, number of searches, mixed and partial profiles/random match probability, presence of family members) when reporting a DNA database match. An example of such a warning can be found in Appendix 3.

**ENFSI recommendation 25**

DNA database managers should be aware of the possibility of adventitious matches and be able to calculate their expected numbers for the matches they report. (A warning can be included in a report, indicating the factors that increase the possibility of an adventitious match such as size of the database, number of searches, mixed and partial profiles/random match probability, presence of family members, etc.).

<sup>62</sup> A.J. Hopwood et al (2012) Science and Justice 52, 185-190. Consideration of the probative value of single donor 15-plex STR profiles in United Kingdom populations and its presentation in United Kingdom courts.

<sup>63</sup> Tvedebrink T, Eriksen PS, Curran JM, Mogensen HS, Morling N. (2012) Analysis of matches and partial-matches in a Danish STR data set. Forensic Sci Int Genet. 6(3): 387-92.



To compare theoretical numbers of adventitious matches with actual ones, a DNA database manager should record adventitious matches and the conditions under which they were found (size of the database, number of searches, etc.) for future analysis, as Tvedebrink et al. have done<sup>64</sup>.

Special attention must be paid to the occurrence of false positive matches when performing large-scale international comparisons of DNA profiles, such as those based on the EU-Prüm decision<sup>65</sup>. As some countries have expressed discontent with the 6 loci rule (a minimum of 6 loci must be matching)<sup>66</sup>, the Next Generation Prüm project, among other goals, aims to discuss the necessity for changes to the matching rules (increase the lower limit to 8 matching loci) in order to decrease the frequency of false positive matches<sup>67</sup>. However, because there are also true 6 and 7 locus matches (which can be found by additional DNA testing), it is being recommended that the matching rules remain as they are, and let each country decide what to do with 6 and 7 locus matches according to their own internal criminal justice requirements and processes.

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<sup>64</sup> Ibid.

<sup>65</sup> Forensic DNA Profiles Crossing Borders in Europe (Implementation of the Treaty of Prüm). Profiles in DNA 2011.

<sup>66</sup> [http://www.europarl.europa.eu/RegData/etudes/STUD/2018/604971/IPOL\\_STU\(2018\)604971\\_EN.pdf](http://www.europarl.europa.eu/RegData/etudes/STUD/2018/604971/IPOL_STU(2018)604971_EN.pdf), Section 4.3

<sup>67</sup> <http://data.consilium.europa.eu/doc/document/ST-12275-2018-INIT/en/pdf>

## 8 Reporting results

Matches in DNA databases are often so-called “cold hits”, which means that there was no prior evidence suggesting that the match would occur. Even in cases where there is prior evidence, this is not usually known to the DNA database manager. This means that reporting should be done in such a way that does not create misconceptions in the mind of the person receiving the match report.

Apart from reporting a match between two DNA profiles (which may contain different loci) as a fact, the match probability or the likelihood ratio of the corresponding loci/alleles should be reported, to provide the person receiving the report with an idea of the evidential value of the match. Because the present kits produce DNA profiles with random match probabilities which are difficult to comprehend for lay people, Hopwood et al.<sup>68</sup> have recommended the use of maximum likelihood ratios for reporting the weight of the evidence for a fully matching 15-plex DNA profile, shown in Table 8.

Relationship	Likelihood ratio
No relationship (random match probability)	1 in 10 <sup>9</sup>
First cousin	1 in 10 <sup>9</sup>
Half-sibling or uncle/nephew	1 in 10 <sup>9</sup>
Parent or child	1 in 10 <sup>7</sup>
Full-sibling	1 in 10 <sup>5</sup>

*Table 8: Proposed maximum likelihood ratios for reporting the weight of the evidence for a fully matching 15-plex DNA profile*

The evidential value of matches with mixed profiles should be reported as a likelihood ratio, which is the ratio of the probability of the results given two alternative propositions: 1) the crime stain contains DNA from the suspect and an unknown unrelated person and 2) the crime stain contains DNA from two unknown, unrelated persons (see also § 3.9 and recommendation 12). The reported LR is only valid for the evaluated propositions and should be recalculated if alternative propositions are put forward.

There has been discussion in the literature and in courts about the appropriate way to report the evidential value of DNA database search results<sup>69,70,71</sup>. In essence, the difference between the evidential value of a DNA match obtained through a DNA database search and a DNA match obtained through comparison with a single suspect lies in other evidence available in the case: with a “cold hit”, other incriminating evidence against the matched person may be completely missing, whereas the comparison of a single suspect is necessarily based on other incriminating evidence. As argued in Sjerps and Meester<sup>72</sup>, the report should therefore contain a warning concerning the possibility of adventitious matches, as mentioned in recommendation 21. This warning should make clear that adventitious matches are possible, and that this possibility should be taken into account, especially when the database match was obtained from a search with a partial DNA profile and in situations where other incriminating evidence is missing or weak. Meester and Sjerps<sup>73</sup> have suggested the inclusion of a table in the match report, which describes the relation between the prior probability and the posterior probability, given the match probability of the match, to help jurors determine the evidential value of the match. An alternative option, which is currently used by the Netherlands Forensic Institute, is to include a special textbox in the match report, which explains the possibility of adventitious matches (see Appendix 3).

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<sup>68</sup> A.J. Hopwood et al (2012) *Science and Justice* 52, 185-190. Consideration of the probative value of single donor 15-plex STR profiles in United Kingdom populations and its presentation in United Kingdom courts.

<sup>69</sup> Kaye, DH. (2009) Rounding up the usual suspects: a legal and logical analysis of DNA database trawls, *North Carolina Law Review* 87: 425-503.

<sup>70</sup> Gittelsohn S. et al. (2012) The database search problem: A question of rational decision making. *Forensic Sci Int.* 222, 186-199

<sup>71</sup> Biedermann A. et al. (2012) A Bayesian network approach to the database search problem in criminal proceedings. *Investigative Genetics* 3, 16

<sup>72</sup> Sjerps, M. & R. Meester (2009). Selection effects and database screening in forensic science. *Forensic Sci Int.* 192 (2009), 56–61.

<sup>73</sup> Meester, R. and Sjerps, M. (2004) Why the effect of prior odds should accompany the likelihood ratio when reporting DNA evidence; *Law, Probability and Risk*, 3, 51-62.

**ENFSI recommendation 26** A DNA database match report of a crime scene-related DNA profile with a person should be informative. It may include an indication of the evidential value of the match (RMP/LR), a warning indicating the possibility of adventitious matches (as mentioned in recommendation 25), and the implication that the match should be considered together with other evidence.

## 9 DNA database software<sup>74</sup>

Software programs designed for the storage and comparison of DNA profiles are referred to as DNA database software. Some programs also have other functions. DNA database software can either be internally developed by a country to meet its own specific needs, or it can be obtained from a developer, who provides it without cost or offers it on a commercial basis. Examples of DNA database programs which can be obtained without cost are:

- CODIS, which has been developed by the FBI for the USA, but which is also available for non-USA law enforcement organizations. A private company (DHA, formerly Leidos) runs a well-organized and skillful helpdesk and computer-based training is available. CODIS has three levels of storing and comparing DNA profiles: local, state and national, which can be used to combine data if there is more than one DNA database in a country (e.g. Spain).
- STR-lab, a program developed in South Africa: <http://strlab.co.za/>

Programs which are or have been commercially available are:

- FSS-iD™, from the former Forensic Science Service in the United Kingdom
- Dimensions, from the Austrian company Ysselbach Security Systems
- eQMS::DNA, from the Croatian company Pardus ([www.Pardus.hr](http://www.Pardus.hr))
- fDMS-STRdb, distributed by the Czech Republic company Forensic DNA Service (<http://dna.com.cz/files/file/fdms-strdb.pdf>)
- *RapidDNA* from the Australian company Forensics International (<http://www.rapiddna.biz>)
- SmallPond (<http://www.smallpondllc.com/>)
- Bode Match (<http://www.bodecellmark.com/pages/bode-match>)

DNA database programs should comply with national personal data protection guidelines, especially those dealing with data quality, integrity and security.

Argentina has developed its own DNA database program called GENis. (<http://www.fundacionsadosky.org.ar/genis/>). However, it is competing with CODIS<sup>75,76</sup>.

One company has launched a cloud-based DNA database specifically for local law enforcement agencies to archive, search and reference DNA profiles from crime scene samples<sup>77</sup> more easily. It remains to be seen if this storage method will be acceptable to the authorities responsible for DNA testing and/or the data protection authorities.

Table 9 shows which DNA database programs are used by different European countries and some international organizations.

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<sup>74</sup> The mentioning of trade names does not mean that ENFSI recommends or endorses any of these programs. The aim of ENFSI is to provide insight into what is available on the market.

<sup>75</sup> <https://www.pagina12.com.ar/108891-un-lobby-contra-el-csi-argentino>

<sup>76</sup> <https://diariosanrafael.com.ar/que-es-codis-el-software-que-el-gobierno-importo-del-fbi-y-se-aplicara-en-mendoza-114718/>

<sup>77</sup> <http://www.sorensonforensics.com/forensics-lab-forensic-dna-testing/dna-forensics-lab-news-forensic-lab-development/sorenson-forensics-launches-new-cloud-based-database-to-simplify-crime-scene-case-management-archival-of-dna-profiles?category=press+releases>

<b>Country</b>	<b>DNA database program</b>
Albania	CODIS
Armenia	No DNA database yet
Austria	Self-developed program
Belgium	CODIS
Bosnia & Herzegovina	CODIS
Bulgaria	Self-developed program
Croatia	CODIS
Cyprus	Self-developed program
Czech Republic	CODIS
Denmark	Self-developed program + CODIS
Estonia	CODIS
Finland	CODIS
<del>Former Yugoslavian Republic of North</del> Macedonia	eQMS::DNA
France	CODIS + Self-developed program
Germany	Self-developed program
Georgia	CODIS
Greece	CODIS
Hungary	CODIS
Iceland	CODIS
Ireland	CODIS
Italy	CODIS
Kosovo	CODIS
Latvia	CODIS
Liechtenstein	Included in the Swiss DNA database
Lithuania	CODIS
Luxembourg	Self-developed program
Malta	CODIS
Montenegro	CODIS
Netherlands	CODIS
Northern Ireland	Self-developed program
Norway	CODIS
Poland	CODIS
Portugal	CODIS
Romania	CODIS
Russia	No DNA database yet
Scotland	Self-developed program
Serbia	Self-developed program
Slovakia	CODIS
Slovenia	Self-developed program
Spain	CODIS
Sweden	CODIS
Switzerland	CODIS
Turkey	No DNA database yet
Ukraine	Self-developed program
United Kingdom (England, Wales, Scotland, North Ireland) <sup>78</sup>	Self-developed program
INTERPOL	Self-developed program
Prüm Treaty countries (exchange database)	Self-developed program or CODIS
ICMP	Self-developed program

*Table 9: DNA database programs used by different European countries and some international organizations.*

<sup>78</sup> Northern Ireland and Scotland have their own DNA databases, even though their profiles are also loaded to the UK National DNA Database.

## 10 Data integrity control measures

For forensic reasons, and in accordance with personal data protection legislation, DNA profiles and their associated information should be entered and stored correctly. For this reason, the manual entry of DNA profiles should be avoided. If this is not possible, DNA profiles should be entered using the double-blind method<sup>79</sup>. A reliable professional database program should be used, with proper logging of all actions and secure ways of importing the DNA profiles, as indicated in § 4.3. Access to the DNA database should be limited by physical and organizational methods to those persons who require access for various reasons (data entry, searching, etc.). Regular back-ups should be made, stored in a safe place and recovered at regular intervals to simulate recovery from a disaster. If the DNA profiles and/or the information associated with DNA profiles are also registered in another system, like a LIMS or a judicial or police system, the contents of these systems should be regularly compared to verify whether the systems are properly synchronized.

Official recognition of compliance with personal data protection legislation may be sought by submitting the organization and its work procedures to an independent, external audit.

### **ENFSI recommendation 27**

- DNA profiles should be entered into a database in a way that guarantees correct entry.
- Access to the DNA database should be limited to those persons who require access, by physical and organizational measures.
- Regular back-ups should be made, stored in a safe place, and recovered at regular intervals to simulate recovery from a disaster.
- When DNA profiles and their associated information are present in different systems, these systems should be regularly compared to verify whether they are properly synchronized.

The above-mentioned recommendations are made to maximize the reduction of errors. It has been shown, however, that despite all of these measures, DNA profiles may still occasionally contain errors as a result of:

- Allele drop-ins or drop-outs
- Allele calling errors (of long DNA fragments)
- Primer mutation differences between commercial kits
- Mixture interpretation errors by DNA analysts

When searching at moderate stringency (see §5.2), DNA profiles containing allele drop-outs and primer mutation differences will appear as a match between a heterozygote and an apparent homozygote, but DNA profiles containing other types of errors will not match their correct counterparts. To detect these false negative matches or false exclusions (e.g. true matches that are not found due to an error in one of the DNA profiles), regular full DNA database searches, allowing one or more mismatches, should be performed, as indicated and recommended in §5.2. The software used by countries exchanging DNA profiles under the terms of the EU Prüm Decision allows for one mismatch. When a match between two DNA profiles contains a mismatch at one of the loci, the original data of both DNA profiles should be checked for any errors.

If no error is found in either profile, it must be concluded that the mismatch is a true mismatch. During the international exchange of DNA profiles based on the EU Prüm Council decisions, many 6 and 7 locus matches plus a mismatch are found. Nearly all of these mismatches have proven to be true mismatches, and statistical calculations also show that these high numbers of 6 and 7 locus matches plus a mismatch are to be expected. Some countries, therefore, have chosen to ignore these matches, except for those which may assist in solving serious cases.

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<sup>79</sup> The double blind method is also used for changing passwords. A new password is entered twice while only asterisks are shown. The computer compares the two blind entries and only accepts it if both entries are equal.

## **11 Inclusion of case information and personal data**

In some countries, the DNA database program also contains case and personal information, but in other countries, this is strictly separated for legislative or other reasons. The DNA database program CODIS has only been developed to store and compare DNA profiles, so CODIS-using countries always need a second system to store other information associated with the DNA profile. As indicated in the previous chapter, regular comparisons of the systems are then required to verify whether they are still properly synchronized, and if the DNA profiles are correctly linked to their associated personal and/or case information.

Whether or not the DNA profiles are kept separated from personal data, the identity of persons should be properly verified when they are sampled to avoid matches with wrong or non-existing persons.

## 12 Interaction with other databases

It can be very useful for investigative reasons to combine DNA information with other technical or tactical forensic information. If, for example, a series of crimes has been linked by the presence of a DNA profile to an unknown person and one of the crime scenes has a fingerprint matching a known person, the combined information may solve the whole series of cases. Countries like the United Kingdom<sup>80</sup>, Switzerland<sup>81</sup> and the Netherlands<sup>82</sup> are working on systems to combine the contents of different forensic databases and to visualize the links between different cases and different persons which result from that combination. Figure 2 shows an example of how the visualization of the links between a cluster (or clusters) of crimes and persons derived from DNA and fingerprint information can be charted.

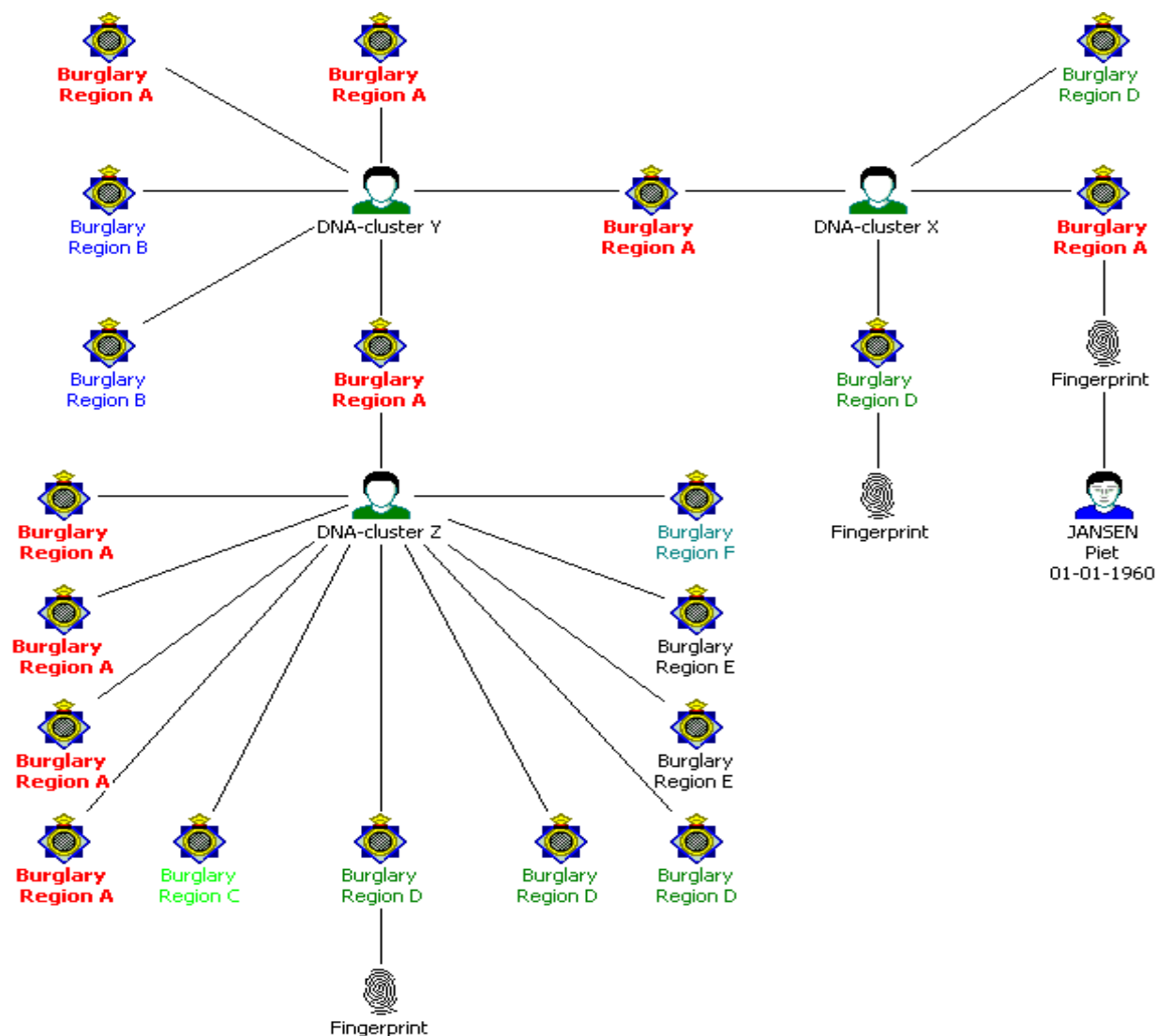


Figure 2: Three DNA clusters (X, Y, Z) linked by 2 crime scenes where DNA profiles from 2 clusters were found (X+Y and Y+Z) combined with two unidentified and one identified fingerprints.

**ENFSI recommendation 28** Investigating authorities should consider combining the information from a national DNA database with other types of evidence to increase the likelihood of identifying leads in other crimes.

<sup>80</sup> <http://webarchive.nationalarchives.gov.uk/+http://www.homeoffice.gov.uk/docs2/resconf2002/richardlearyrolenimflints.pdf>

<sup>81</sup> Ribaux et al. (2010) Intelligence-led crime scene processing. Part II: Intelligence and crime scene examination. Forensic Science International 199 (1-3) 63-71

<sup>82</sup> See English summary of: [http://www.wodc.nl/images/1203\\_volledige%20tekst\\_tcm44-58753.pdf](http://www.wodc.nl/images/1203_volledige%20tekst_tcm44-58753.pdf)



## 13 Automation of work processes

Automation of DNA database work processes can take place at different levels:

- Import of DNA profiles as already discussed in § 4.3
- Comparison of DNA profiles using saved sets of matching rules
- Comparison of DNA profiles at scheduled points in time (e.g. overnight)
- Reporting unambiguous results
- Sending out the unambiguous results

As automated processes reduce the possibility of human error, they should be introduced for those processes that are straightforward, like the production of DNA profiles from reference samples.

As already stated in § 2.7, candidate matches with mixed profiles should always be checked by a DNA expert to determine whether the numerical match could be a true match. This is also the reason why mixed DNA profiles are not included in the automated DNA comparisons between countries operating under the terms of the EU Prüm Decision.

Recently, so-called rapid DNA machines have come on the market, for the fully automated production of DNA profiles from reference samples in less than 2 hours. Once these machines have proven themselves, they may alter the DNA testing landscape, because their use would enable quick DNA testing, conducted quickly, on location at police stations, to verify the identities of arrestees or suspects within the DNA database before their release. In the USA, a mega project is going on to realize this<sup>83</sup>. However, some concerns have been raised in the implementation of this technology<sup>84, 85</sup>. The field is evolving quickly, and efforts are being made into the development of crime-stain-appropriate rapid DNA machines as well, with varied success<sup>86</sup>. Nevertheless, it should be noted that in Europe, for example, there are binding quality assurance requirements in regular DNA laboratories (EN/ISO 17025), as well as country-specific legislation, that may prohibit the use of this technology in the forensic area. The use of this rapid DNA technology for crime scene stain analyses is currently not recommended, as it could lead to the destruction of irretrievable important biological stain material.

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<sup>83</sup> <https://www.fbi.gov/services/laboratory/biometric-analysis/codis/rapid-dna>

<sup>84</sup> <https://www.latimes.com/california/story/2019-09-24/rapid-dna-forensics-crime-police>

<sup>85</sup> L. Wilson-Wilde, F. Pitman, Legislative and policy implications for the use of Rapid DNA technology in the Australian context, *Forensic Sci. Policy Manage.* (2017), <http://dx.doi.org/10.1080/19409044.2017.1335809>

<sup>86</sup> Boiso et al. (2017) RapidHIT for the purpose of stain analyses – An interrupted implementation. *Forensic Science International: Genetics Supplement Series* 6 e589–e590

## 14 Storage of cell material

The cell material of crime scene stains from which a DNA profile has been generated is usually stored. Regarding the storage of cell material from reference samples, however, different countries have different policies. Some countries allow the storage of reference samples for later reuse, if this becomes technically or legally necessary, while in other countries, the reference samples must be destroyed as soon as the DNA profile has been generated and included in the DNA database. The following three examples show that, from a forensic point of view, it is better to store the cell material.

### Example 1

In the recent past, several improved DNA typing technologies have been developed. Multiplex kits with more loci for higher evidential value and higher sensitivity, as well as mini-STR kits and SNP kits to obtain DNA profiles from degraded DNA, are good examples. It has become possible to re-examine stains from (c)old cases that could not be examined in the past. But if the stain has been retyped with new technology, the reference sample must also be retyped, to enable a more stringent comparison between the two. If the reference sample has been destroyed, the police or the judiciary must obtain a new reference sample from the suspect, which may not always be possible.

### Example 2

A Prüm Treaty member country sends an SGM+ DNA profile of a crime scene stain to another Prüm Treaty member country. A match with a reference DNA profile is reported for 7 loci due to the fact that the matching country uses a different kit. To exclude the possibility of an adventitious match, the SGM+ country first tries to improve its own DNA profile, but if this is not possible, it requests that the matching country upgrade its reference DNA profile. If the reference sample has been destroyed, this upgrade is not possible without obtaining a new reference sample from the person involved, which may not always be possible.

### Example 3

Countries which are allowed to perform familial searching in their DNA database usually get many possible candidates after an initial DNA database search. False positives can be eliminated from this possible candidate list by additional Y-chromosomal or mitochondrial DNA testing. However, this is only possible if the samples from which the DNA profiles were generated are still available.

The ENFSI DNA Working Group realizes that the storage of cell material from reference samples is politically a very delicate subject. Although the European personal data protection directive clearly states that personal data (which includes DNA profiles and the cell material from which the DNA profiles are derived) can only be used for the purpose for which they were obtained, there are people who fear that they could be misused in the future and hence choose the “better safe than sorry” principle and choose to destroy the sample after a profile has been included in the DNA database. These concerns have not been reduced with the implementation of GDPR and LED legislation in May of 2018.

On the other hand, one could also argue that keeping the samples enhances privacy, because there is no need for resampling if additional DNA testing is necessary to investigate a possible false positive match to determine whether it is a true match.

### **ENFSI recommendation 29**

If possible, the cell material of reference samples should be stored to permit further processing, such as a loci upgrade, depending on internal laboratory procedures or national legislation.

## 15 Legislative matters

As the compulsory taking of a DNA sample is a breach of an individual's privacy and bodily integrity, Article 8 of the European Convention on Human Rights demands justification and legislation. For arrestees and suspects, justification is found in the fact that DNA testing can help solve a case by either finding a match (resulting in possible incriminating evidence) or an exclusion (resulting in possible exonerating evidence) with a DNA profile from a crime scene thought to be left behind by the culprit of the crime. This means, however, that crime scene DNA must be present for this type of justification. The inclusion of an individual's DNA in a DNA database is justified by the fact that it can help solve old and future crimes committed by the same person, and that it may prevent new crimes because the person involved may fear being detected. The indefinite retention of a person's DNA profile in a DNA database without prosecution or conviction has been condemned by the European Court of Human Rights<sup>87</sup>. The Court has explicitly approved the retention of the DNA of innocent people in appropriate circumstances by praising the Scottish retention system. Also, in the Netherlands, suspects can be kept in the DNA database until their case has been dealt with by the public prosecution office. Similarly, in Latvia, the data of a person with the status of suspect in a specific case must be deleted from the DNA database if the case against them has been closed on rehabilitative grounds, or if they have been found not guilty in court, but only if there are no other active cases against them. Their data will also be retained if they have been convicted in the past.

Every EU country should have data protection legislation derived from Regulation (EU) 2016/679 (General Data Protection Regulation) or, depending on the status of the institution conducting DNA analysis, Directive (EU) 2016/680 (Law Enforcement Directive). Because DNA profiles and the cell material from which they are derived are also regarded as personal data, they fall under the umbrella of this legislation, unless the data protection legislation is overruled by specific DNA legislation containing other provisions (*Lex Specialis* precludes *Lex Generalis*). Some examples are given below to illustrate why it is useful to have specific DNA legislation in addition to data protection legislation:

- According to data protection legislation, personal data must not be stored longer than is necessary for the purpose for which it was collected. It is practically impossible to determine this necessity at regular intervals for all the DNA profiles in a DNA database. Therefore, DNA legislation provides guidelines on storage times (see also: § 3.1).
- According to data protection legislation, individuals have certain rights with regard to their own data (access/modification/removal). For investigative reasons, this is usually not desirable. Therefore, DNA legislation states who has access to information present in, and generated by, the DNA database.
- In some countries, data protection legislation states that genetic information can only be used in relation to the person from whom this information is derived. If such a country would allow familial searching in the DNA database, appropriate rules for this should be provided in the DNA legislation.

DNA profiles are not only very specific for an individual, but they also contain information about the relatives of that individual. This means that, when people voluntarily give their DNA profile (e.g. in a mass screen), they should be informed that this may possibly incriminate a relative. In this way, a person can decide whether they will make use of their right not to testify against relatives.

Most countries also allow the inclusion of DNA profiles from minors in their DNA database. The legitimacy of this is under question in some countries, with reference to the international convention on the rights of the child. Several appeal court cases are ongoing to develop jurisprudence on this. The Supreme Court of the Netherlands has ruled that there is no reason to differentiate between minors and adults. Additionally, the European Court of Human Rights does not regard minority as a reason to exclude a person from the Dutch DNA database<sup>88</sup>.

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<sup>87</sup> <http://www.bailii.org/eu/cases/ECHR/2008/1581.html>

<sup>88</sup> European Court on Human Rights 20689/08.

## 16 Financing

In most countries, the costs of establishing and maintaining a National DNA database are financed by a dedicated annual budget under the Ministry of Justice or the Ministry of the Interior. In the United Kingdom, however, (parts of) the budget are managed by the police, who pay for the production and the storage of the DNA profiles.

## 17 Personnel requirements

It goes without saying that personnel working with any DNA database should be properly trained to use the DNA database software. If the program is self-developed, this will be in-house training. If the DNA database software is commercially obtained, the company selling the software will usually offer training in the use of the software. CODIS can be obtained from the FBI by law enforcement organizations, and computer-based training is available.

Apart from undergoing proper training, DNA database personnel must have the following personal skills as a minimum:

- Ability to work very conscientiously
- Ability to keep confidential information confidential
- Ability to accept verification by colleagues
- Ability to report own mistakes to enable further process improvement

Apart from the above-mentioned requirements, a “proof of good conduct” may be required, sometimes even a positive outcome in an investigation by the police or the secret service into a candidate’s reliability.

## 18 Governance

When a DNA database is established in a country, its custody is either assigned to an existing organization, or to a newly established organization. In some countries, a special supervisory board has been established, staffed with the representatives of different stake-holders. A list of such bodies can be found at: <http://www.cfbdadosadn.pt/en/conexoes/adndireitos/Pages/outrosconselhosfiscalizacao.aspx>. In the United Kingdom, a special ethics group has been established<sup>89</sup> to provide independent advice on the ethical aspects of DNA database management. If there is no dedicated supervisory board, the data protection authority of a country usually has the power to audit the organization managing the DNA database, to verify its compliance with the data protection legislation of that country.

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<sup>89</sup> See: <https://www.gov.uk/government/organisations/national-dna-database-ethics-group>

## 19 Research and Development

Studies on the statistics, performance and different search strategies of DNA databases are usually done using simulated DNA databases (e.g. footnotes 5 and 24). Some scientists, however, have asked for disclosure of the actual DNA profiles contained in DNA databases, to allow them to evaluate some of the assumptions in population genetics underlying DNA testing<sup>90</sup>. Of course, this should be done under strict conditions and by removing any links to the identity of the owner of the DNA profile. Some countries already allow this in the interest of quality assurance and/or process improvement<sup>91,92,93</sup>.

A major issue for DNA database managers is that they cannot distinguish matches between monozygotic twins. Both epigenetic<sup>94</sup> as well as next generation sequencing<sup>95</sup> research is occurring, but the amounts of DNA which are necessary for these analyses must be reduced to enable analysis of forensic traces containing low amounts of DNA.

Social scientists also study DNA databases. A recent review was produced by the EUFORGEN project<sup>96</sup>. The European Horizon 2020 project “EXCHANGE” has recently begun and will run until 2020<sup>97</sup>.

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<sup>90</sup> Krane et al. (2009) *Science* 326,1631-1632. Time for DNA-disclosure.

<sup>91</sup> Sjerps et al (2009) Oral presentation at the European Academy of Forensic Science (EAFS) 2009 conference in Glasgow. Observed and expected numbers of (partially) randomly matching profiles in the Dutch DNA-database and in international searches.

[http://dnadatabank.forensischinstituut.nl/Images/eafs-2009-glasgow-presentatie-marjan-sjerps\\_tcm127-477445.ppt](http://dnadatabank.forensischinstituut.nl/Images/eafs-2009-glasgow-presentatie-marjan-sjerps_tcm127-477445.ppt)

<sup>92</sup> Tvedebrink et al (2012) *Forensic Science International: Genetics* Volume 6, Issue 3 , Pages 387-392, May 2012. Analysis of matches and partial matches in a Danish STR data set.

<sup>93</sup> Hedell et al. (2011) *Forensic Science International: Genetics Supplement Series* Volume 3, Issue 1, Pages e135-e136, December 2011.

<sup>94</sup> Li et al (2011) *Forensic Science International: Genetics Supplement Series* Volume 3, Issue 1 , Pages e337-e338, December 2011.

Identical but not the same: The value of DNA methylation profiling in forensic discrimination within monozygotic twins.

<sup>95</sup> J. Weber-Lehmann et al (2014) *Forensic Science International: Genetics* 9, 42-46. Finding the needle in the haystack: Differentiating “identical” twins in paternity testing and forensics by ultra-deep next generation sequencing

<sup>96</sup> [http://www.eurofor-gen.eu/fileadmin/websites/eurofor-gen/images/Dissemination\\_Documents/WP4/Williams\\_and\\_Wienroth\\_-\\_2013\\_-\\_Systematic\\_Review.pdf](http://www.eurofor-gen.eu/fileadmin/websites/eurofor-gen/images/Dissemination_Documents/WP4/Williams_and_Wienroth_-_2013_-_Systematic_Review.pdf)

<sup>97</sup> [http://dnadatabank.forensischinstituut.nl/Images/project-description-from-ces-newsletter\\_tcm127-609633.pdf](http://dnadatabank.forensischinstituut.nl/Images/project-description-from-ces-newsletter_tcm127-609633.pdf)

## 20 External Communication

Because DNA databases are usually publicly funded, politicians, the public and the media have a right to know how the DNA database is managed and what results are obtained.

### 20.1 Annual report

A good way to make this information publicly available is to produce an official annual report. Such a report can either be part of the annual report of the organization responsible for the management of the national DNA database, or it can be a separate annual report dedicated solely to the DNA database. In Europe, the United Kingdom, Sweden, Belgium and the Netherlands have already produced such dedicated annual reports. Outside Europe, the Royal Canadian Mounted Police also produces an annual report for their DNA database. Below are the locations where the most recent issues of these annual reports can be downloaded:

- United Kingdom: <https://www.gov.uk/government/collections/dna-database-documents>
- Netherlands: [http://dnadatabank.forensischinstituut.nl/dna\\_dossier/jaarverslagen\\_dna\\_databank/index.aspx](http://dnadatabank.forensischinstituut.nl/dna_dossier/jaarverslagen_dna_databank/index.aspx)
- Canada: <http://www.rcmp-grc.gc.ca/nddb-bndg/index-accueil-eng.htm>
- Belgium: <https://nicc.fgov.be/upload/images/ODcriminalistiek/dna-databank-jv-pub-nl-screen.pdf> and <https://nicc.fgov.be/upload/images/ODcriminalistiek/dna-databank-jv-pub-fr-screen.pdf>
- Sweden: <http://www.nfc.polisen.se/Global/www%20och%20Intrapolis/Statistik/Statens%20kriminaltekniska%20laboratorium/%C3%85rsrapport%20dna-register%202015.pdf?epslanguage=Automatisk>

### 20.2 Internet site

Whereas annual reports are milestones in written form, websites provide a continuous way of providing information to interested parties. Below is a list of internet sites devoted to DNA databases, or containing information about DNA databases:

#### Europe

- United Kingdom: <https://www.gov.uk/government/collections/dna-database-documents>
- Germany: [https://www.bka.de/nn\\_205980/DE/ThemenABisZ/DnaAnalyse/dna.html#doc205380bodyText5](https://www.bka.de/nn_205980/DE/ThemenABisZ/DnaAnalyse/dna.html#doc205380bodyText5)
- Ireland: [http://www.lawreform.ie/\\_fileupload/Reports/Report%20DNA%20Database.pdf](http://www.lawreform.ie/_fileupload/Reports/Report%20DNA%20Database.pdf)  
(comprehensive thoughts on setting up a DNA database in Ireland)
- Netherlands: <http://dnadatabank.forensischinstituut.nl/>
- Switzerland: <https://www.fedpol.admin.ch/fedpol/en/home/sicherheit/personenidentifikation/dna-profile.html>
- Portugal: <http://www.cfbdadosadn.pt/en>
- Belgium: <https://nicc.fgov.be/nationale-dna-databanken> and <https://incc.fgov.be/banques-nationales-de-donnees-adn>
- Sweden: <http://nfc.polisen.se/kriminalteknik/biologi/dna-register/>
- Spain: [http://www.interior.gob.es/documents/642317/1203227/Base\\_de\\_datos\\_policia\\_identificadores\\_AD\\_N\\_126190539\\_web.pdf/](http://www.interior.gob.es/documents/642317/1203227/Base_de_datos_policia_identificadores_AD_N_126190539_web.pdf/)

#### Rest of the world

- USA (CODIS) <http://www.fbi.gov/about-us/lab/codis/codis>
- USA (Florida): [http://www.fdle.state.fl.us/Content/getdoc/6835b26c-ae3f-49c5-845e-0c697bb86001/DNA\\_Investigative.aspx](http://www.fdle.state.fl.us/Content/getdoc/6835b26c-ae3f-49c5-845e-0c697bb86001/DNA_Investigative.aspx)
- USA (New York): <http://criminaljustice.state.ny.us/forensic/dnabrochure.htm>
- USA (Legislation): <http://www.dnaresource.com/>
- Canada: <http://www.rcmp-grc.gc.ca/nddb-bndg/index-accueil-eng.htm>
- Australia: [http://www.crimtrac.gov.au/our\\_services/BiometricServices.html](http://www.crimtrac.gov.au/our_services/BiometricServices.html)
- New Zealand: <http://www.esr.cri.nz/forensic-science/our-work/dna-databank/>



➤ Hong Kong: [http://www.govtlab.gov.hk/english/abt\\_fsd\\_dds.htm](http://www.govtlab.gov.hk/english/abt_fsd_dds.htm)

## 21 International overviews

Several documents have been published in the past containing country-specific overviews on the different aspects of DNA database legislation and DNA database management. However, most of these documents are now older than 5 years.

Two more recent country-specific overviews are presently known to the author:

- One can be found at the website of the Council of Responsible Genetics: [http://www.councilforresponsiblegenetics.org/dnadata/world\\_map.html](http://www.councilforresponsiblegenetics.org/dnadata/world_map.html); however, not all content is correct or up-to-date, as the report was issued in 2011
- The other has been produced by the EuroforGen project in 2014, with an update in 2016, and an upcoming update in 2019/2020: [https://www.euroforGen.eu/fileadmin/websites/euroforGen/images/Dissemination\\_Documents/WP4/Reed\\_and\\_Syndercombe\\_Court\\_2016\\_Legal\\_Audit.pdf](https://www.euroforGen.eu/fileadmin/websites/euroforGen/images/Dissemination_Documents/WP4/Reed_and_Syndercombe_Court_2016_Legal_Audit.pdf)

## 22 International comparison of DNA profiles

As crimes committed in one country may be committed by a person from another country, it is very useful to have the means for the international comparison of DNA profiles. Chapter 2 contains descriptions of how a European Standard Set of Loci has been agreed upon to enable such comparisons. In addition to common loci, countries exchanging DNA profile information should, of course, also use the same quality standards for the production of their DNA profiles, as described in § 3.5.

There are different channels through which the international comparison of DNA profiles can take place:

- Individual legal assistance requests on paper  
For the majority of countries outside of Prüm (see below), this is still the most commonly used channel. Depending on the legal embedding of the DNA legislation of a country, either police channels or judicial channels are used for this method of exchanging single search requests of DNA information. Before the advent of XML to communicate DNA profiles, INTERPOL developed a special form to standardize and facilitate this manner of exchanging DNA information. Since 2012, an electronic version has been available in all countries, where applicants complete the form (either typing in data or by XML upload) and send the request to their choice of selected countries, and/or to the INTERPOL DNA database. A great disadvantage of this manner of information exchange is that it is very time-consuming.
- INTERPOL DNA database and DNA gateway  
In 2002, INTERPOL created a central DNA database, in which DNA profiles and their sample codes can be included for comparison by its 190 member states. The database is an autonomous database and does not keep any nominal data linking a DNA profile to any individual. Member States retain the ownership of their profile data and control its submission, access by other countries and destruction in accordance with their national laws. When a match is found, a message is immediately sent to the countries contributing to the match. This message contains the basic case information that was provided, and can optionally provide the sample codes. Member countries then decide if they wish to pursue this forensic intelligence link. A central DNA database is most effective when all participating countries submit all their crime scene DNA profiles and all their reference sample DNA profiles. There are currently 81 participating countries providing DNA profiles in accordance with their national laws. To encourage further participation, an INTERPOL resolution was adopted by all countries at the 78th INTERPOL General Assembly for National Central Bureaus to facilitate the submission of DNA profiles from unsolved crime scenes and foreign national offenders by national law enforcement agencies to the INTERPOL DNA database (<http://www.interpol.int/News-and-media/Events/2009/78th-INTERPOL-General-Assembly2/78th-INTERPOL-General-Assembly-Resolutions>).

The INTERPOL DNA Gateway is a medium for the transfer of DNA profiles between two or more countries, and for the management of a country's own DNA profiles in the central DNA database. Access to the DNA gateway is provided directly to a country via INTERPOL National Central Bureaus (NCBs), using INTERPOL's secure communications system, I-24/7. For more information about the DNA gateway of INTERPOL, see: [https://www.interpol.int/download/file/FS01\\_02\\_2015\\_EN\\_web](https://www.interpol.int/download/file/FS01_02_2015_EN_web). Interpol has also published a second (2009) edition of the Interpol Handbook on DNA Data Exchange: <http://www.interpol.int/content/download/8993/66934/version/6/file/HandbookPublic2009.pdf>. The results of a Global Survey from 2016 can also be viewed from their DNA information page: <https://www.interpol.int/How-we-work/Forensics/DNA> <sup>98</sup>

In addition to the regular DNA gateway facilities, INTERPOL offers the possibility of dedicated bilateral comparisons using the INTERPOL DNA Bilateral Matcher.

- Europol

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<https://www.interpol.int/content/download/4875/file/INTERPOL%20Global%20DNA%20Survey%20Results%202016%20%28Public%20Version%29.pdf>

Europol is authorized to process DNA profiles within the “Analysis Work Files” (Council Decision 2009/9343/JHA). In this context, DNA profiles are used together with other intelligence for criminal analysis purposes in order to fight serious international crime. Europol can also process DNA profiles in the Europol Information System (EIS) based on Council Decision 2009/371/JHA. Europol National Units can directly insert or search data, including DNA profiles, in the EIS. Non-EU states which have signed a co-operation agreement with Europol can also provide DNA profiles to Europol for insertion into the EIS. Bulgaria has begun the addition of their Prüm match results to the EIS.

- The EU Prüm Decisions (derived from the Treaty of Prüm)

The EU Prüm Decisions deal with the exchange of judicial and police information between EU Member States. Some associated countries (Norway, Switzerland, Liechtenstein and Iceland) have also been permitted to join this undertaking. With regards to DNA, countries can search each others’ DNA databases in an automated way. To enable this, each country creates a copy of its DNA database with a standardized table structure, which can be accessed by common data exchange and DNA comparison software, present in each country. The DNA data exchange and matching system used by EU Member States is similar to the DNA data exchange and matching system of the INTERPOL DNA Gateway.

The EU Prüm Decision (2008/615/JHA) and the EU Prüm Implementation Decision (2008/616/JHA) can be found at the following internet locations:

<http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2008:210:0001:0011:EN:PDF>

<http://eur-lex.europa.eu/LexUriServ/LexUriServ.do?uri=OJ:L:2008:210:0012:0072:EN:PDF>

In 2019, the following countries are already exchanging DNA profiles on a day-to-day basis with one or more of the other countries, under the terms of the EU Prüm Decisions:

Austria,  
Belgium,  
Bulgaria,  
Croatia,  
Cyprus,  
Czech Republic,  
Denmark.  
Estonia,  
Finland,  
France,  
Germany,  
Hungary,  
Ireland,  
Latvia,  
Lithuania,  
Luxembourg,  
Malta,  
Netherlands,  
Poland,  
Portugal  
Romania,  
Slovakia,  
Slovenia,  
Spain,  
Sweden,  
United Kingdom.

These countries have updated their exchange software to enable the exchange of the 5 new ESS-loci, in addition to other loci used in each country’s DNA database.

Chapter 2 of the Appendix to the EU Prüm Implementation Decision contains the DNA inclusion, matching and reporting rules. However, due to the advances and developments in the field of forensic DNA analysis, these rules are undergoing review<sup>99</sup>.

Like the INTERPOL DNA database, the Prüm DNA profile exchange system is a match/no-match system, meaning that only DNA profiles are compared. After finding a match, countries can obtain the personal and/or case information associated with the DNA profile via existing police or judicial channels. The minimum number of matching loci under the terms of the Prüm system is six. However, it can be calculated, and it has been shown in daily practice, that six and seven locus matches have a non-negligible chance of being false positive<sup>100</sup>. Therefore, it is recommended that these matches are analysed further by additional DNA testing before any legal action is undertaken against any matching person.

**ENFSI recommendation 30** If a Prüm-related information request is received from another country, the quality of the corresponding match should be verified before providing the requested information to the other country.

**ENFSI recommendation 31**

If possible, when operational under the Prüm treaty, six- and seven-locus international matches should be further analyzed by additional DNA testing before requesting information from another country.

The minimum number of loci for a DNA profile to participate in the Prüm system is six, and the required minimum number of matching loci is also six. One should realize, however, that two profiles which both fulfil the inclusion rule may not match if there is not enough overlap to produce the minimum number of six matching loci. This is especially true for over 600.000 “old” German reference profiles which consist of the old 7 ESS loci + SE-33.

To improve the evidential value of a match by additional DNA testing, one must know which loci are present in the DNA profile of the other country. Therefore, those loci that are not used by the receiving country should be configured in the DNA database of the receiving country. If this is not done, those loci will not be visible in the DNA profile received from the sending country.

**ENFSI recommendation 32**

All regularly-used loci (in addition to those not used by the receiving country) should be configured in the DNA databases of countries participating in the international exchange of DNA profiles under the terms of the Prüm system in order to see the full composition of the DNA profile of the sending country.

Countries that compare DNA profiles under the terms of the EU Prüm decisions should regularly perform 3 checks:

- Check if all profiles that comply with the Prüm inclusion rules are Prüm-marked
- Check if all Prüm-marked profiles have been sent to all active Prüm labs
- Check if an answer was received and processed from all active Prüm labs for every profile that was sent out

A Power Point presentation on how to perform these checks in CODIS can be downloaded from the European CODIS Users Platform at Europol

<sup>99</sup> <http://data.consilium.europa.eu/doc/document/ST-12275-2018-INIT/en/pdf>

<sup>100</sup> Forensic DNA Profiles Crossing Borders in Europe (Implementation of the Treaty of Prüm). Profiles in DNA 2011 (<http://www.promega.com/resources/articles/profiles-in-dna/2011/forensic-dna-profiles-crossing-borders-in-europe/>).

- The PCSC Agreements  
The USA has negotiated so called PCSC (Prevention and Combatting Serious Crime) agreements with several European countries. A PCSC agreement resembles the Prüm agreement and provides for the reciprocal exchange of biometric and biographic data and any relevant underlying information for law enforcement purposes. Some countries already compare fingerprints with the USA on the basis of such a treaty but the DNA exchange has not yet come into force because the federal DNA law of the USA has to be adjusted to give other countries access to the DNA database of the USA.
- PCC SEE (Police Cooperation Convention for South East Europe)  
PCC SEE is a cooperation between Albania, Austria, Bosnia and Herzegovina, Bulgaria, Hungary, Macedonia, Moldavia, Montenegro, Romania, Serbia and Slovenia. These countries have decided to set up a Prüm-like system<sup>101</sup>. Austria, Bulgaria, Hungary, Romania and Slovenia are already Prüm countries, so the other countries can benefit from their experience. The European Commission has accused these 4 countries of infringing on the exclusive competence of the EU in this area ([https://ec.europa.eu/commission/presscorner/detail/en/INF\\_19\\_5950](https://ec.europa.eu/commission/presscorner/detail/en/INF_19_5950)).

A national DNA database always contains DNA profiles from national crime-related stains. However, as already mentioned in § 3.1, if the national law of both countries allows it, crime-related stains from other countries may also be included, if an international legal request for comparison from another country has not resulted in a match. By including the DNA profile from the other country, there is no need for a regular repeat of the often lengthy legal request. This is, of course, not necessary for countries that already search each others' DNA databases under the terms of the EU Prüm Decision, because the request can be repeated as frequently as needed.

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<sup>101</sup> <https://www.pccseesecretariat.si/libs/download.php?file=/8dba58dc1687894a70673cbc0122ff11> and <https://www.pccseesecretariat.si/libs/download.php?file=/8a621f1388a219ba63188c9a1e2bd620>

## 23 Missing persons

### 23.1 Introduction

The main purpose of a DNA database for missing persons is to see whether the DNA profiles of unidentified human remains can be linked to DNA profiles of missing persons or their family members. In this way, the family members of missing persons can be made aware of the fact that their missing relative is no longer alive and can start coming to terms with their loss. A second purpose is to link the body parts of a single person, which may be found in different locations (e.g. two feet washed ashore at different places and at different times), or in situations where more than one person has been killed and unidentified body parts cannot be reliably attributed to one person (e.g. airplane crash or secondary mass grave). Missing persons DNA databases are usually operated together with, or as part of, a system where other important attributes of missing persons and unidentified bodies/body parts can also be included and compared (e.g. dental records, fingerprints, externally visible traits, medical data, etc.). Samples obtained from the personal items of missing persons or samples obtained from their family members are indicated as *ante mortem* samples, and a sample from an unidentified body (or body part) is indicated as a *post mortem* sample.

### 23.2 Different missing person situations

A person can become a missing person either as an individual or as part of a mass fatality. Mass fatalities may have a natural cause (e.g. tsunami, earthquake) or a human-induced cause (e.g. war situation, terrorist attack). In mass fatalities, a distinction can also be made between closed systems, where the number, names and mutual relationships of the missing persons are known (e.g. airplane crash) and open systems, where the number of missing persons cannot be properly assessed (e.g. tsunami or earthquake).

### 23.3 Different types of matches

In the DNA databases of missing persons, a distinction can be made between direct and indirect comparisons.

- A direct match is either a full match between the DNA profile of a missing person and the DNA profile of an unidentified body(part), or a full match between different body parts.
  - A direct match between the DNA profile of a missing person and the DNA profile of an unidentified body(part) is the most reliable type of match, but it requires that the DNA profile of the missing person be available – that it was obtained, for instance, from a personal item or a medical specimen of the missing person. Care should be taken to ensure that the personal item was used only by the missing person. To verify this, DNA profiles should also be obtained from the parents or children of the missing person, for comparison with the DNA profile obtained from the personal item. The strength of a direct match is usually expressed as the random match probability of the matching loci between the DNA profile of the missing person and the DNA profile of the unidentified body(part), or as a likelihood ratio expressing the probability of the results, given the following two propositions: either the DNA is from the missing person, or it is from an unknown person.
  - Sometimes different body parts of an unidentified person are found. These can then be linked to each other by direct matches. DNA database programs may include the possibility to combine these profiles into one so-called representative specimen without losing the original data. This should only be done if the Likelihood Ratio of the individual profiles is high enough to allow for this kind of clustering.
- An indirect comparison is a comparison between the DNA profiles of persons that are possibly related to the missing person. For instance, to investigate whether an unidentified body(part) of a particular missing person, the DNA profile can be compared to the DNA profiles of the missing person's relatives. This approach is used when the DNA profile of the missing person is not available for direct comparison. In this case, the strength of the match is usually expressed as a likelihood ratio (e.g. assuming that X is indeed the biological child of Y and Z, the result of the DNA analysis is x times more likely if the unidentified body(part) is X than if it is a random unrelated person). Specialized software is available to perform these calculations (see § 23.6). Some of these programs (like CODIS and Bonaparte) have the possibility to build pedigrees and add DNA data to the nodes of a pedigree

by the drag-and-drop principle. The use of prior odds for missing persons identifications has been discussed by Budowle et al.<sup>102</sup> and Thompson et al.<sup>103</sup>.

Compared to forensic DNA testing, the identification of missing persons or the victims of disasters is even more complex. There may be inconsistencies in the reference pedigree due to unknown relationships. Additionally, mutations and partial profiles may cause problems and/or false positive and false negative matches. Also, the statistics are more complex when compared to forensic DNA testing. Therefore, additional training is necessary for DNA experts involved in kinship testing. This has been confirmed by a collaborative exercise of the Spanish- and Portuguese-speaking Working Group of the ISFG<sup>104</sup>.

Kinship (pedigree) searches can be conducted in CODIS as well as other database programs.

## **23.4 Markers**

A comparison between the DNA profiles in a missing persons DNA database usually starts with 10-15 autosomal STR markers. In the case of a direct match, the evidential value of the match will usually be sufficient for the decision-maker to identify the person, but in the case of an indirect match, additional autosomal markers may have to be determined, as well as Y-STR markers and/or mtDNA, to verify or falsify the match.

## **23.5 Relationship between criminal and missing persons DNA databases**

In some countries, DNA profiles of missing persons (and/or their relatives) and unidentified human remains are kept in the same DNA database as the DNA profiles used for solving crimes, while in other countries a separate DNA database is used for missing persons (and/or their relatives) and unidentified human remains. There may be several reasons for this:

- Data protection considerations. By keeping DNA profiles of missing persons and their relatives separate from the DNA profiles in the criminal DNA database, they cannot be accidentally compared with profiles with which they should not be compared;
- Both DNA databases may be managed by different organizations (e.g. Ministry of Justice versus the Police);
- Specialized software is needed to find and evaluate matches between unidentified human remains and multiple relatives in pedigrees of missing persons.

If two separate DNA databases are used, it must be kept in mind that it can be useful to compare the DNA profiles of unidentified human remains with the DNA profiles of the criminal DNA database:

- DNA profiles of unidentified human remains found in one location may match with stains found at a crime scene at another location, indicating that the unidentified person may have been the victim of a crime (if this was not yet obvious) and has been transported to another location;
- DNA profiles of unidentified human remains may match with a reference sample, which may assist an identification. This comparison needs to be done only once, as the unidentified person is dead and hence cannot be added to the DNA database as a reference sample in the future.

## **23.6 Software**

Specialized software is available to search for relatedness between (a series of) DNA profiles and/or to calculate the likelihood ratio of the relatedness of a person and their putative family member(s). This type of software is also used in forensic and civilian cases to verify or falsify the biological relationship between known persons. Table 10 lists the different programs that are known to the editors of this document<sup>105</sup>.

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<sup>102</sup> Budowle et al (2011) Investigative Genetics 2: 15. Use of prior odds for missing persons identifications

<sup>103</sup> Thompson et al. (2013) Frontiers in Statistical Genetics and Methodology 4(220), pp. e1-e3, 10-2013. The role of prior probability in forensic assessments.

<sup>104</sup> Vullo et al. (2016) GHEP-ISFG collaborative simulated exercise for DVI/MPI: lessons learned about large scale profile database comparisons. Forensic Sci. Int. Genet. 21: 45-53

<sup>105</sup> The mentioning of trade names does not mean that ENFSI recommends or endorses any of these programs. The aim of ENFSI is to provide insight into what is available on the market.



Program	Developer	Country	Website	Price	Remarks
Bloodhound	Ananomouse Corporation	USA	<a href="http://www.ananomouse.com/products/bloodhound.asp">http://www.ananomouse.com/products/bloodhound.asp</a>	Unknown	URL does not refer to the software
Bonaparte Disaster Victim Identification System	SMART Research	Netherlands	<a href="http://www.bonaparte-dvi.com/">http://www.bonaparte-dvi.com/</a>	Not free	
CODIS 7.0	FBI	USA	<a href="https://www.fbi.gov/about-us/lab/biometric-analysis/codis">https://www.fbi.gov/about-us/lab/biometric-analysis/codis</a>	Free	Only for Law enforcement organizations
DNASat	Jaroslav Berent	Poland	<a href="http://www.umed.lodz.pl/ou/zms/">http://www.umed.lodz.pl/ou/zms/</a>	Unknown	URL does not refer to the software
DNA-View	Charles Brenner	USA	<a href="http://dna-view.com/index.html">http://dna-view.com/index.html</a>	Not free	
EasyDNA	Wing Kam Fung	Hong Kong	<a href="http://www.hku.hk/statistics/EasyDNA/">http://www.hku.hk/statistics/EasyDNA/</a>	Free	Accompanying book is essential
EasyPat	Michael Krawczak	Germany	<a href="http://www.uni-kiel.de/medinfo/mitarbeiter/krawczak/download/">http://www.uni-kiel.de/medinfo/mitarbeiter/krawczak/download/</a>	Free	
familias	Petter Mostad	Norway	<a href="http://www.familias.name/">http://www.familias.name/</a>	Free	
FSS DNA Lineage	Forensic Science Service	UK	<a href="http://www.forensic.gov.uk/html/services/analytical-solutions/software/fssibd/">http://www.forensic.gov.uk/html/services/analytical-solutions/software/fssibd/</a>	Unknown	URL does not refer to the software
GeneMarker HID	SoftGenetics	USA	<a href="http://www.softgenetics.com/GeneMarkerHID.html">http://www.softgenetics.com/GeneMarkerHID.html</a>	Not free	
GenomiCalc	Genomic	Brazil	<a href="http://www.genomicalc.com.br">http://www.genomicalc.com.br</a>	Unknown	URL does not refer to the software
GenoProof2	Qualitytype	Germany	<a href="http://qualitytype.de/genoproof/">http://qualitytype.de/genoproof/</a>	Not free	
Genolab	Qualitytype	Germany	<a href="http://www.genolab.eu">http://www.genolab.eu</a>	Not free	
Genotype	Kvant	Slovakia	<a href="http://www.dip.sk/typo3/dip.sk/index.php?id=9&amp;no_cache=1&amp;L=1">http://www.dip.sk/typo3/dip.sk/index.php?id=9&amp;no_cache=1&amp;L=1</a>	Unknown	URL does not refer to the software
Grape	DNA-SOFT	USA	<a href="http://www.dna-soft.com">http://www.dna-soft.com</a>	Not free	
Hugin	Hugin Expert	Denmark	<a href="http://www.hugin.com/products/services/demo/hugin-lite">http://www.hugin.com/products/services/demo/hugin-lite</a>	Not free	URL does not refer to the software
KIn CALc	California DOJ/Steven Myers	USA	<a href="mailto:Steven.Myers@doj.ca.gov">Steven.Myers@doj.ca.gov</a>	Unknown	No URL
KINGROUP	Dmitry A. Kononov	Australia	<a href="http://www.kingroup.org/">http://www.kingroup.org/</a>	Unknown	URL does not refer to the software
LISA	Future Technologies, Inc.	USA	<a href="http://www.ftечи.com/dna_biometric.shtml">http://www.ftечи.com/dna_biometric.shtml</a>	Not free	
M-FISys	Gene Codes Forensics	USA	<a href="http://www.genecodesforensics.com/software/">http://www.genecodesforensics.com/software/</a>	Not free	
PatCan	Jose Antonio Riancho	Spain	<a href="mailto:jose.riancho@unican.es">jose.riancho@unican.es</a>	Unknown	No URL
Patern	Michael Krawczak	Germany	<a href="http://www.uni-kiel.de/medinfo/mitarbeiter/krawczak/download/">http://www.uni-kiel.de/medinfo/mitarbeiter/krawczak/download/</a>	Free	
Paternity Index	Michael Jung	Germany	<a href="http://www.paternityindex.com/">http://www.paternityindex.com/</a>	Not free	
PatPCR	Juan Antonio Luque	Spain	<a href="mailto:vestad@telepolis.com">vestad@telepolis.com</a>	Unknown	No URL
PedExpert	Sérgio Danilo Junho Pena	Brazil	<a href="mailto:spena@dcc.ufmg.br">spena@dcc.ufmg.br</a>	Unknown	No URL
REPAIR	William L. Duren, Michael Epstein, Mingyao Li, and Michael Boehnke	USA	<a href="http://csg.sph.umich.edu/boehnke/repair.php">http://csg.sph.umich.edu/boehnke/repair.php</a>	Free	
SmallPond	SmallPond LLC	USA	<a href="http://www.smallpondllc.com/">http://www.smallpondllc.com/</a>	Not free	
VAT	Max Baur, Rolf Fimmers, W. Spitz	Germany	<a href="http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4025154/">http://www.ncbi.nlm.nih.gov/pmc/articles/PMC4025154/</a>	Not free	URL does not refer to the software

Table 10: Software programs to search for relatedness between (a series of) DNA profiles and/or to calculate the likelihood ratio of the relatedness of a person and their putative family member(s).

Depending on the required application of the software, different program properties will be more or less important to have. Table 11 lists some program parameters which should be considered when choosing (buying) a missing persons software program.

Parameter category	Parameter
Data which can be compared	Autosomal STRs
	Y-STRs
	mtDNA
	SNPs
	Metadata
Search strategies	UHR against UHRs
	UHRs against UHRs to find relations
	UHR against MPs
	UHR against pedigrees of MP
	UHRs against pedigrees of MP
	Familial searching (shared alleles)
	Familial searching (LR-ranking)
Calculations	Pedigree likelihood ratio calculation
	Fst correction
	Size-bias correction
	Mutation correction <sup>106</sup>
	Allele drop-out correction
	Multiple allele relative frequency tables
	Minimum allele relative frequency substitution for rare alleles
	Datafilters
	Resultfilters
Other features	Graphical pedigree manager
	Combining DNA profiles of the same person
	Incestuous relationships
	Reporting module
	Simulations module
	Import module
	Replacing MP by IP in pedigree

Table 11: Program parameters which may be considered when choosing (buying) a missing persons software program (UHR: Unidentified Human Remains; MP: Missing Person; IP: Identified Person; LR: Likelihood Ratio)

Because software programs are continuously adapted and improved, interested persons should refer to the producer of the program to find out the latest properties.

<sup>106</sup> A recent discussion about different mutation models can be found in: Chakraborty et al (2011) Investigative Genetics 2:8. Response to: DNA identification by pedigree likelihood ratio accommodating population substructure and mutations- authors' reply.

## **23.7 International Organisations**

### **23.7.1 International Commission on Missing Persons (ICMP)<sup>107</sup>**

The International Commission on Missing Persons was established at the initiative of U.S. President Clinton in 1996 at the G-7 Summit in Lyon, France. Its primary role is to ensure the co-operation of governments in locating and identifying those who have disappeared during armed conflict or as a result of human rights violations. ICMP also supports the work of other organizations, encourages public involvement in its activities and contributes to the development of appropriate expressions of commemoration and tribute to the missing.

The organization was established to support the Dayton Peace Agreement, which ended the conflicts in Bosnia and Herzegovina. ICMP is currently headquartered in Sarajevo. In addition to its work in the countries of former Yugoslavia, ICMP is now actively involved in helping governments and other institutions in various parts of the world address social and political issues related to missing persons and establish effective identification systems in the wake of conflict or natural disaster.

Since November 2001, ICMP has led the way in using DNA as a first step in the identification of large numbers of persons missing from armed conflict. ICMP has developed a DNA database of over 90,000 relatives of 29,500 missing people, and more than 54,000 bone samples taken from mortal remains exhumed from clandestine graves in the countries of former Yugoslavia. By kinship matching of STR profiles from bone samples and family references, ICMP has been able to identify over 17,000 people who were missing from the conflicts and whose mortal remains were found in hidden graves. Additionally, ICMP provides extensive assistance in similar post-conflict or human-rights-related missing persons efforts in Latin America, Africa, the Mediterranean and the Middle East.

ICMP has been involved in a number of large-scale DVI efforts, including the 2004 SE Asian tsunami, the 2006 Hurricane Katrina in the United States and the 2008 Typhoon Frank in the Philippines. ICMP has established agreements to partner efficiently with INTERPOL, DVI operations in particular, to provide DNA testing and matching capabilities, and to participate in INTERPOL's Incident Response Teams (IRTs) that are often deployed upon invitation by the relevant national authorities to assess and help guide DVI response activities.

ICMP has developed a comprehensive Forensic Data Management System (fDMS) that permits data tracking and analysis for an integrated identification system that includes forensic archaeology, forensic anthropology, missing persons and relatives, DNA matching, and reporting. The fDMS can be flexibly modified to various contexts, and includes a web-based Online Inquiry Center that aids in the establishment of missing persons databases and provides an information link for data to both the general public and partnering forensic authorities, as deemed appropriate for the context and particular roles. Central to ICMP's role in maintaining large databases of potentially sensitive information are ICMP's data protection policies and recognized privileges and immunities that ensure data protection both in policy and practice.

### **23.7.2 INTERPOL**

INTERPOL member countries can call for assistance in disaster victim identification (DVI).

The services offered by INTERPOL include:

- A downloadable DVI guide with Ante Mortem and Post Mortem report forms available on the INTERPOL public website;
- Assistance from the Command and Co-ordination Centre at the INTERPOL General Secretariat in Lyon, France, to send messages between National Central Bureaus 24 hours a day in Arabic, English, French or Spanish;

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<sup>107</sup> The text of this paragraph was supplied by ICMP

- An Incident Response Team to provide further assistance upon request, such as on-site investigative support or connection to INTERPOL's databases.

After the 2004 Tsunami in Thailand, an EU research and development project to develop an international 'Missing Persons/Unidentified Dead Bodies' database using primary and secondary identifiers was funded by the EU's 7th Framework Program. This three-year project, named FASTID, was completed in March 2013 upon the finalization of a prototype database. In order to proceed with the next phase of integrating the MPUB prototype into the INTERPOL General Secretariat, a proposal has been submitted to the EU for funding.

INTERPOL has tested the use of the Bonaparte software for the international comparison of DNA profiles of unidentified persons and the family members of missing persons and is planning to offer this service to its member countries by the end of 2017 <sup>108</sup>.

INTERPOL also has a central DNA database at its General Secretariat in Lyon, which is described in chapter 20 of this document. In this DNA database, the DNA profiles of missing persons and unidentified bodies can also be included and compared. The missing persons index is only compared against the unidentified human remains index. The following countries have submitted missing persons profiles and/or unidentified human remains profiles to the INTERPOL DNA Database:

Algeria	Gibraltar	Poland
Austria	Greece	Portugal
Australia	Hong Kong	Romania
Azerbaijan	Hungary	Russian Federation
Belarus	India	Serbia
Belgium	Iraq	Slovakia
Bosnia & Herzegovina	Ireland	Slovenia
Brazil	Israel	South Africa
Bulgaria	Italy	Spain
Canada	Japan	Sweden
Chile	Jordan	Switzerland
Costa Rica	Kuwait	Syrian Arab Republic
Croatia	Latvia	Thailand
Cyprus	Liechtenstein	The Netherlands
Czech Republic	Lithuania	Turkey
Denmark	Luxembourg	Ukraine
Estonia	Mexico	United Arab Emirates
Finland	Montenegro	United Kingdom
France	New Zealand	UN Mission in Kosovo
North Macedonia	Norway	United States
Germany	Panama	

### **23.8 European missing persons DNA databases**

The table below, which is based on the INTERPOL Global DNA profiling 2014 Survey and on directly obtained information, contains an inventory of countries in Europe which have the DNA profiles of missing persons and unidentified human remains in their DNA databases.

<sup>108</sup> <http://www.interpol.int/News-and-media/News/2013/PR141>

<b>Country</b>	<b>Missing Persons and Unidentified Human</b>	<b>Separated from (S) or included (I) in the criminal DNA-database</b>	<b>Software</b>
Albania	?	?	?
Andorra	Yes	?	?
Austria	Yes	I	National design
Belgium	No	n/a	n/a
Bosnia & Herzegovina	?	?	?
Bulgaria	Yes	I	National Design
Croatia	Yes	I	CODIS
Cyprus	Yes	I	National Design
Czech Republic	Yes	I	CODIS
Denmark	Yes	I+S	CODIS + Plassdata
Estonia	Yes	I	CODIS
Finland	Yes	I	CODIS
North Macedonia	No	n/a	n/a
France	Yes	S	CODIS
Germany	Yes	S	National design
Georgia	Yes	?	?
Greece	Yes	?	?
Hungary	Yes	I	CODIS
Iceland	No	n/a	n/a
Ireland	Yes	?	?
Italy	Yes	I	National Design
Latvia	Yes	I	CODIS
Liechtenstein	Yes	I	CODIS
Lithuania	Yes	I	CODIS
Luxembourg	No	n/a	n/a
Malta	Yes	?	?
Montenegro	Yes	?	?
Netherlands	Yes	S	Bonaparte; CODIS; DNAView
Northern Ireland	?	?	?
Norway	Yes	I	CODIS
Poland	Yes	I	CODIS
Portugal	Yes	n/a	n/a
Romania	Yes	I	CODIS
Russia	Yes	I	National Design
Scotland	?	?	?
Serbia	No	?	?
Slovakia	Yes	I	CODIS
Slovenia	Yes	I	National Design
Spain	Yes	S	CODIS
Sweden	No	n/a	n/a

Switzerland	Yes	?	CODIS
Turkey	Yes	?	?
Ukraine	No	n/a	n/a
United Kingdom (England + Wales)	Yes	S	?

*Table 12: Missing persons DNA databases in Europe (? = not known to the author; n/a = not applicable)*

## **Appendix 1: Summary of ENFSI recommendations on DNA database management**

- 1) Every EU/ENFSI country should establish a forensic DNA database and pass specific legislation for its implementation and management.
- 2) The type of crime-related stain DNA-profiles which can be included in a DNA database should not be restricted.
- 3) To increase the chance of identifying the donors of stains, the number of persons in a DNA database who are likely to be the donors of those stains should be as large as legally (and financially) possible.
- 4) Managers of national DNA databases should establish (together with other stake-holders) criteria for the inclusion of partial DNA profiles to obtain an acceptable balance between the minimum allowable level of evidential value (maximum random match probability) of a DNA profile and the maximum number of adventitious matches a partial DNA profile is expected to generate.
- 5) If possible, DNA profiles should be upgraded after a match in the national DNA database if it increases the evidential value of the match and decreases the possibility of an adventitious match.
- 6) Reference sample profiles should preferentially be loaded to a database only if a complete profile (maximum number of loci) is obtained using the PCR chemistry of choice.
- 7) Labs producing DNA profiles for a DNA database should, as a minimum, be ISO-17025 (and/or nationally equivalent) accredited and should participate in challenging proficiency tests.
- 8) The custodian of the DNA database should have regular contacts with the suppliers of the DNA profiles to exchange information about legal and technical developments, changes in the inclusion and matching rules, incidents, etc.
- 9) If a laboratory uses enhanced techniques to produce DNA profiles they should be searched using a dedicated (near) match strategy.
- 10) Composite DNA profiles should only be created from DNA profiles generated from the same DNA extract because it cannot be excluded that different extracts, even from the same sample, contain DNA from different individuals.
- 11) When a new allele is observed in a DNA profile, its presence should be confirmed by repeated DNA extraction, PCR, capillary electrophoresis and allele calling of the entire DNA profile. Only new alleles whose size can be accurately determined using the internal DNA size-standard should be included in the DNA database.
- 12) Alleles from loci with chromosomal anomalies may be included in a DNA database if the default search strategy is “moderate”. If the default search strategy is “high”, wild cards may be used, as long as an agreed set of wildcards is determined to permit meaningful international exchange.
- 13) The guidelines in the document of the ISFG working group on the analysis of mixed profiles should be used for the analysis of mixed profiles. Software tools may also be used, provided they are properly validated.
- 14) A numerical match between a reference sample and a mixed profile must always be checked against the electropherogram of the mixed profile.
- 15) Mixed profiles of more than 2 individuals should not be systematically included in a DNA database because they will generally produce too many adventitious matches.
- 16) Databases may contain autosomal STR profiles only. For those databases containing profiles from non-autosomal STR profiles or mitochondrial DNA sequences, specific operating procedures must be in place to avoid unintended familial searches. To avoid false exclusions, clear rules should be in place to indicate differences between a mtDNA sequence and the rCRS when comparing mtDNA results.

- 17) If the removal of a DNA profile from the DNA database is dependent on external instruction from an authorized agent, a process should be in place to inform the custodian of the DNA database of this instruction, preferably by means of an automated message.
- 18) There should be a system that can be consulted by those responsible for taking reference samples, to verify whether a person is already present in the DNA database.
- 19) DNA databases should contain an associated elimination DNA database (or databases). This should include laboratory staff of all categories, as well as visitors and maintenance personnel and profiles from those with access to traces (e.g. police, crime scene technicians).
- 20) Because elimination databases are not shared with other EU/ENFSI countries, unidentified DNA profiles found in negative controls, which may originate during the manufacture of disposables and/or chemicals should be uploaded to the ICMP Manufacturers Exclusion Database, MED.
- 21) Policies and procedures should be in place to ensure that DNA-profiles deemed no longer relevant by the authorizing agent are deleted.
- 22) The occurrence of errors in DNA profiles as a result of human mistakes associated with data entry should be avoided as much as possible by automating the allele calling and the DNA database import process. Automated processes reduce the possibility of human error, however, when DNA profiles are entered manually into the DNA database, a process that detects typing errors, for example the double-blind method of entry, should be used.
- 23) To prevent and detect false exclusions (e.g. true matches that are not found due to an error in one of the DNA profiles), DNA profiles should be searched using a full Database search allowing at least one mismatch. The original data of DNA profiles involved in such near matches should be checked for possible errors during their production and processing.
- 24) As a national DNA database is regularly subject to attention from the public, politicians and the media, a DNA database manager should consider establishing tools to monitor the effectiveness of their DNA database and communicating this objective information publicly.
- 25) DNA database managers should be aware of the possibility of adventitious matches and be able to calculate their expected numbers for the matches they report. (A warning can be included in a report, indicating the factors that increase the possibility of an adventitious match such as size of the database, number of searches, mixed and partial profiles/random match probability, presence of family members, etc.).
- 26) A DNA database match report of a crime scene-related DNA profile with a person should be informative. It may include an indication of the evidential value of the match (RMP/LR), a warning indicating the possibility of adventitious matches (as mentioned in recommendation 25), and the implication that the match should be considered together with other evidence.
- 27) DNA profiles should be entered into a database in a way that guarantees correct entry. Access to the DNA database should be limited to those persons who require access, by physical and organizational measures. Regular back-ups should be made, stored in a safe place, and recovered at regular intervals to simulate recovery from a disaster. When DNA profiles and their associated information are present in different systems, these systems should be regularly compared to verify whether they are properly synchronized.
- 28) Investigating authorities should consider combining the information from a national DNA database with other types of evidence to increase the likelihood of identifying leads in other crimes.
- 29) If possible, the cell material of reference samples should be stored to permit further processing, such as a loci upgrade, depending on internal laboratory procedures or national legislation.
- 30) If a Prüm-related information request is received from another country, the quality of the corresponding match should be verified before providing the requested information to the other country.
- 31) If possible, when operational under the Prüm treaty, six- and seven-locus international matches should be further analyzed by additional DNA testing before requesting information from another country.



**32)** All regularly-used loci (also those not used by the receiving country) should be configured in the DNA databases of countries participating in the international exchange of DNA profiles under the terms of the Prüm system in order to see the full composition of the DNA profile of the sending country.

## Appendix 2: ENFSI guidelines for auditing DNA databases

This appendix document aims to provide practical guidelines for teams auditing a DNA database with the intention of verifying its compliance with the ENFSI DNA Working Group recommendations. The document also aims to provide a reporting format for the auditing team, which can be filled out at the auditing site and can be presented to the person(s) requesting the audit. The recommendations of the ENFSI DNA Working Group, as listed in Appendix 1, have been taken as the foundation of the auditing operation, and the opinion of the auditor can be added to each item.

In 2008, the Council of the European Union agreed on converting major parts of the Treaty of Prüm into two EU Council decisions (2008/615/JHA and 2008/616/JHA). These decisions describe the obligation for EU Member States to establish a DNA database, and to make it available for automated searches by other Member States.

Any Member State initiating data exchange after 13 October 2009 must also pass an evaluation procedure (6661/2/09 Rev 2) consisting of:

- Filling out a questionnaire on data protection (6661/1/09 Rev 1 Add 1 Rev 1);
- Filling out a questionnaire on the exchange of DNA profiles (6661/1/09 Rev 1 Add 2 Rev 1);
- A pilot run to test and validate the IT environment;
- An evaluation visit by an external evaluation team to verify all the information provided;
- The approval of the EU Council based on the report of the evaluation team.

The EU Working Party on Data Protection and Information Exchange (DAPIX) has developed guidelines and a reporting format for the evaluation teams. Although there is some overlap between the guidelines of the EU and ENFSI, their focus is quite different. The ENFSI guidelines focus on the proper functioning and management of a DNA database in a national environment, while the EU guidelines focus on the interaction of a DNA database with other DNA databases and on their compliance with the contents of the two EU Council decisions (2008/615/JHA and 2008/616/JHA). Together, they offer an instrument to determine proper management in a national, as well as an international, environment.

It should be noted that ENFSI has established its recommendations based on forensic optimization criteria. Sometimes the national legislation is in contradiction with the ENFSI recommendations. In such cases, the auditor can indicate a Noncompliance with ENFSI guidelines, but that this Noncompliance is acceptable because national law supersedes the ENFSI guidelines.

# GENERAL INFORMATION

Country	
Audit date(s)	
Audit requested by	
Hierarchy of the database	national/sub-national
Organizational position of the database	
Auditing persons (function)	
Database manager(s)	
Database user(s)	
Database IT personnel	
Sources of DNA profiles	

# AUDIT QUESTIONS

ENFSI recommendation 1	Every EU/ENFSI country should establish a forensic DNA database and pass specific legislation for its implementation and management.
Audit question(s)	Are copies of the legislation available (or internet sources where they can be found available)?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 2	The type of crime-related stain DNA profiles which can be included in a DNA database should not be restricted.
Audit question(s)	What are the criteria for the inclusion of stains?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 3	To increase the chance of identifying the donors of stains, the number of persons in a DNA database who are likely to be the donors of those stains should be as large as legally (and financially) possible.
Audit question(s)	What are the criteria for the inclusion of individuals?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 4	Managers of national DNA databases should establish (together with other stake-holders) criteria for the inclusion of partial DNA profiles to obtain an acceptable balance between the minimum allowable level of evidential value (maximum random match probability) of a DNA profile and the maximum number of adventitious matches a partial DNA profile is expected to generate.
Audit question(s)	What are the criteria for the inclusion of partial profiles?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 5	If possible, DNA profiles should be upgraded after a match in the national DNA database if it increases the evidential value of the match and decreases the possibility of an adventitious match.
Audit question(s)	Is it possible to upgrade older/partial profiles? If so what is the reason to update the older/partial profiles?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 6	Reference sample profiles should preferentially be loaded to a database only if a complete profile (maximum number of loci) is obtained using the PCR chemistry of choice.
Audit question(s)	What are the criteria for the inclusion of reference samples?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 7	Labs producing DNA profiles for a DNA database should, as a minimum, be ISO17025 (and/or national equivalent) accredited and should participate in challenging proficiency tests.
Audit question(s)	Which labs produce DNA profiles for the DNA database and are they (in the process of being) accredited?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 8	The custodian of the DNA database should have regular contacts with the suppliers of the DNA profiles to exchange information about legal and technical developments, changes in the inclusion and matching rules, incidents, etc.
Audit question(s)	Does the custodian have regular contacts with the suppliers of the DNA profiles?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 9	If a laboratory uses enhanced techniques to produce DNA profiles they should be searched using a dedicated (near) match strategy.
Audit question(s)	If the lab uses enhancement techniques to produce DNA profiles (do you use a near match search strategy?)
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 10	Composite DNA profiles should only be created from DNA profiles generated from the same DNA extract because it can not be excluded that different extracts, even from the same sample, contain DNA from different sources.
Audit question(s)	Does the DNA database contain composite DNA profiles and if so, how were they created?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	



ENFSI recommendation 11	When a new allele is observed in a DNA profile, its presence should be confirmed by repeated DNA extraction, PCR, capillary electrophoresis and allele calling of the entire DNA profile. Only new alleles whose size can be accurately determined using the internal DNA size standard, should be included in the DNA database.
Audit question(s)	Is there a written procedure for the inclusion of new/rare alleles?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 12	Alleles from loci with chromosomal anomalies may be included in a DNA database if the de-fault search strategy is "moderate". If the default search strategy is "high", wild cards may be used, as long as an agreed set of wildcards is determined to permit meaningful international exchange.
Audit question(s)	Is there a written procedure for the handling of chromosomal anomalies?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

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ENFSI recommendation 13	The guidelines in the document of the ISFG working group on the analysis of mixed profiles should be used for the analysis of mixed profiles. Software tools may also be used, provided they are properly validated.
Audit question(s)	Is there a written procedure for the processing of mixed DNA profiles (both in the lab and in the DNA database)?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 14	A numerical match between a reference sample and a mixed profile must always be checked against the electropherogram of the mixed profile.
Audit question(s)	See the question associated with recommendation 12.
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 15	Mixed profiles of more than 2 individuals should not be systematically included in a DNA database because they will generally produce many adventitious matches.
Audit question(s)	See the question associated with recommendation 12.
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 16	Databases may contain autosomal STR profiles only. For those databases containing profiles from non-autosomal STR profiles or mitochondrial DNA sequences, specific operating procedures must be in place to avoid unintended familial searches. To avoid false exclusions, clear rules should be in place to indicate differences between a mtDNA sequence and the rCRS when comparing mtDNA results.
Audit question(s)	Are non-autosomal STR profiles or mitochondrial profiles added to the criminal DNA database? If yes, are specific operating procedures in place to avoid unintended familial searches? Are clear rules in place to indicate differences between a mtDNA sequence and the rCRS when comparing mtDNA profiles?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 17	If the removal of a DNA profile from the DNA database is dependent on external instruction from an authorized agent, a process should be in place to inform the custodian of the DNA database of this instruction, preferably by means of an automated message.
Audit question(s)	What are the rules and procedures for informing the removal of DNA profiles from the DNA database?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 18	There should be a system that can be consulted by those responsible for taking reference samples, to verify whether a person is already present in the DNA database.
Audit question(s)	Is there a system that can be consulted by those responsible for sampling persons to see whether a person is already present in the DNA database?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 19	DNA databases should contain an associated elimination DNA database (or databases). This should include laboratory staff of all categories, as well as visitors and maintenance personnel and profiles from those with access to traces (e.g. police, crime scene technicians).
Audit question(s)	Do you have an elimination database in place?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 20	Because elimination databases are not shared with other EU/ENFSI countries, unidentified DNA profiles found in negative controls, which may originate during the manufacture of disposables and/or chemicals should be uploaded to the ICMP Manufacturers Exclusion Database, MED.
Audit question(s)	Are the DNA profiles detected in negative controls shared with the MED?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 21	Policies and procedures should be in place to ensure that DNA-profiles deemed no longer relevant by the authorizing agent are deleted.
Audit question(s)	Are there policies and procedures in place?

Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 22	The occurrence of errors in DNA profiles as a result of human mistakes associated with data entry should be avoided as much as possible by automating the allele calling and the DNA database import process. Automated processes reduce the possibility of human error, however, when DNA profiles are entered manually into the DNA database, a process that detects typing errors, for example the double-blind method of entry, should be used.
Audit question(s)	Describe the allele calling and DNA database inclusion process. If not fully automated, which measures have been put in place to avoid human error?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 23	To prevent and detect false exclusions (e.g. true matches that are not found due to an error in one of the DNA profiles), DNA profiles should be searched using a full Database search allowing at least one mismatch. The original data of DNA profiles involved in such near
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	matches should be checked for possible errors during their production and processing.
Audit question(s)	Are DNA profiles checked for mistakes using a near match approach in a whole of database approach?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 24	As a national DNA database is regularly subject to attention from the public, politicians and the media, a DNA database manager should consider establishing tools to monitor the effectiveness of their DNA database and communicating this objective information publicly.
Audit question(s)	Is the performance of the DNA database monitored and communicated to the public?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 25	DNA database managers should be aware of the possibility of adventitious matches and be able to calculate their expected numbers for the matches they report. (A warning can be included in a report, indicating the factors that increase the possibility of an adventitious match such as size of the database, number of searches, mixed and partial profiles/random match probability, presence of family members, etc.).
Audit question(s)	Can the database manager/laboratory calculate the expected number of adventitious matches?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 26	A DNA database match report of a crime scene-related DNA profile with a person should be informative. It may include an indication of the evidential value of the match (RMP/LR), a warning indicating the possibility of adventitious matches (as mentioned in recommendation 25), and the implication that the match should be considered together with other evidence.
Audit question(s)	What does the Database match report include?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	



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ENFSI recommendation 27	DNA profiles should be entered into a database in a way that guarantees correct entry. Access to the DNA database should be limited to those persons who require access, by physical and organizational measures. Regular back-ups should be made, stored in a safe place, and recovered at regular intervals to simulate recovery from a disaster. When DNA profiles and their associated information are present in different systems, these systems should be regularly compared to verify whether they are properly synchronized.
Audit question(s)	Does the laboratory implement these management strategies?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 28	Investigating authorities should consider combining the information from a national DNA database with other types of evidence to increase the likelihood of identifying leads in other crimes.
Audit question(s)	Are you aware of this investigative strategy in your country?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 29	If possible, the cell material of reference samples should be stored to permit further processing, such as
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	a loci upgrade, depending on internal laboratory procedures or national legislation.
Audit question(s)	What are the rules and procedures under the legislation for the destruction of the cell material of reference samples?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 30	If a Prüm-related information request is received from another country, the quality of the corresponding match should be verified before providing the requested information to the other country.
Audit question(s)	Is the match verified before providing the requested information?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 31	If possible, when operational under the Prüm treaty, six- and seven-locus international matches should be
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	further analyzed by additional DNA testing before requesting information from another country.
Audit question(s)	Are six and seven locus matches further analyzed before requesting information?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

ENFSI recommendation 32	All regularly-used loci (also those not used by the receiving country) should be configured in the DNA databases of countries participating in the international exchange of DNA profiles under the terms of the Prüm system in order to see the full composition of the DNA profile of the sending country.
Audit question(s)	Is it possible to show which loci have been configured in your DNA database?
Audit result	
Compliance? If not, why?	Yes / No
Noncompliance acceptable? If yes, why?	Yes / No
Remark(s)/recommendation(s)	

## **Appendix 3: English translation of the textbox included in Dutch match reports**

### **POINT OF ATTENTION WITH REGARDS TO A DNA DATABASE MATCH**

DNA databases contain large numbers of DNA profiles of known persons and of biological traces related to unsolved crimes.

As the number of DNA profiles in a DNA database increases, so does the chance of obtaining an adventitious match with a person who is not the actual donor of the trace.

This is especially true for partial DNA profiles and mixed DNA profiles, because the chance that they would match with a randomly chosen person is greater than the chance that a full, single DNA profile would match a randomly chosen person.

If there are doubts as to whether the matching person is the donor of the trace, for instance - because there is no other tactical or technical evidence which links the person to the crime, the possibility for additional DNA testing should be considered.

This point of attention applies particularly to matches found as the result of large-scale international DNA profile comparisons based on the EU-Prüm decisions.

## Appendix 4: Changes in the 2019 document relative to the 2017 document

- Kit table, one locus removed, other kits verified and updated.
- Recommendations comprehensively reviewed by 21 Database countries (major alterations to Recommendations 5, 6, 9, 12, 16, 17, 19, 20, 21, 22, 23, 24, 25, 26, 28, 29, 30, 31).
- Recommendation 12 reversed to permit uploading of autosomal anomalies, provided moderate search strategy used.
- Audit trail changed to reflect updated recommendations
- Strider Database reference included pt 3.6
- Other providers of mixture interpretations included in pt 3.8 Mixed profiles, along with non-endorsement disclaimer.
- Most recent EMPOP version added to pt 3.12 Mitochondrial DNA (mtDNA) information.
- Paragraph on recent discussion regarding a universal DNA database added pt 3.13 Universal DNA database.
- References and contact information for ICMP MED added in pt 4.5.
- Reference to genealogy databases (commercial) added in pt 5.2 Search modes, footnote regrading GEDMatch and initial US police use of the database.
- Additional search strategies in CODIS added in pt 5.4 Near matches.
- Additional information regarding match rate added, as well as other parameters in pt 6.2.
- Information regarding the comparison of efficiencies of DNA databases removed.
- Table 4: Semi-annual ENFSI DNA database overview for 2018 updated, henceforth will be Annual ENFSI DNA database overview; additional references to studies about DNA database efficiency or effectiveness added to pt 6.2 Output / Efficiency measurement.
- Added information about Next Generation Prüm in Chapter 7 Adventitious matches.
- Added information regarding Argentinian database software in Chapter 9 DNA database software.
- Updated information about the development of rapid DNA technology added to final paragraph in Chapter 13 Automation of work processes.
- Reference to GDPR and LED added in Chapter 14 Storage of cell material and Chapter 15 Legislative matters.
- Interpol links updated in Chapter 22 International comparison of DNA profiles, link added to 2016 Global Survey.
- Reference to Next Generation Prüm added in Chapter 22 International comparison of DNA profiles.
- Comment on European Commission Letter of formal notice to Austria, Bulgaria, Hungary and Romania, including references in footnotes.
- Added paragraph about CODIS pedigree and kinship searches, also reference to other software in pt 23.3 Different types of matches.
- Word 'authors' changed to 'editors of this document' in pt 23.6 Software.
- Update to Table 12: Missing persons DNA databases in Europe.
- Name of Former Yugoslavian Republic of Macedonia updated to North Macedonia throughout document.
- Various spelling, grammar and formatting changes that do not alter the meaning of the text.